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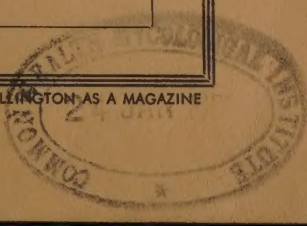

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(AGRICULTURAL RESEARCH SECTION)

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NOTICES

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THE BORDER ROW EFFECT IN WHEAT TRIALS WITH DIFFERENT SPACINGS BETWEEN PLOTS

By JEAN G. MILLER and N. S. MOUNTIER, Extension Division, Department of Agriculture, Wellington

(Received for publication, 9 August 1955)

Summary

The effect on the plot yields of the inclusion of border rows in drilled cereal trials was evaluated in 3 trials where two contrasting varieties were sown at different spacings. Significant differences in plot yields were almost always recorded between the spacings 7 in., 14 in., 18 in., and 28 in., when the outside rows were included. The yields of the second rows from the edge did not vary according to the spacings between plots. There was no clear evidence of a competition effect between varieties, and the effect on the comparative merits of the varieties differed in the 3 trials. No advantage of using 7 in. or 28 in. spacings instead of the standard 14 in. was apparent and these present practical difficulties. Coefficients of variation were increased when border rows were included if the error was calculated to include variations due to different spacings. This corresponds to the case in routine trials where the actual gap between plots varies from 7 to 18 in. Where no spacings differences contribute to error, as in the case of a very accurately drilled trial, the coefficients of variation were not affected by the inclusion of outside rows at the 14 or 18 in. spacings.

The conclusions are that while in some cases whole plots may be harvested, in most cases, where the drilling is not extremely accurate or competition effects may exist, the trial will be improved by the removal of outside rows.

These results apply to variety trials only, and then only to trials where there are no extreme differences of habit or yield between varieties.

INTRODUCTION

In sowing routine cereal trials the standard practice of the Department of Agriculture is to use a field drill with 7 coulters spaced 7 in. apart. Plots are 3 chains by one drill width, and a gap of 14 in. is left between the outside coulters of adjacent plots. The effect of this gap in increasing the growth of the outside rows relative to the others in a plot is often clearly visible and hence it is the practice at harvest to cut these rows separately and discard them when weighing the yield. Where a header harvester is used this doubles the work and will be worthwhile only if:

(a) Outside row effects differ with some variations in spacing.

(b) Outside row effects are greater with some varieties or treatments than others.

(c) A vigorous outside row of a tall or high yielding variety depresses the yield of the adjacent shorter or weaker variety (competition effect).

(d) The coefficient of variation is reduced by eliminating the apparently highly variable outside rows.

This problem of the effect of border rows on trial plots has always been the concern of experimentalists but other studies have not usually tried to measure the effect with deliberately varied spacings.

DETAILS OF TRIALS

To test these hypotheses, three trials were conducted.

Trial Symbol	Year	Place
I1	1952/3	Invermay Agricultural Research Station, (Mosgiel)
I2	1953/4	"
W1	1953/4	Winchmore Irrigation Research Station (Ashburton)

The treatments were:—

(a) spacing between plots 7, 14, 18, and 28 in.

(b) variety.

It was intended that a short and a tall variety should be compared, and for the 1952-3 season Hilgendorf was used as the short variety, and a new high-yielding cross from Crop Research Division known as 313,01 as the tall. However, 313,01 did not grow as tall as expected, and Hilgendorf, being an early variety, was the taller during the early part of the growing season.

In the 1953-4 season 313,01 was used as the short variety and another cross 393,01 as the tall.

The spacings were chosen to cover the likely range of variation (7 in. to 18 in.) in a routine trial, and the 28 in. spacing was added to test the theory that at a very wide distance competition between plots is eliminated and the outside row effect is consequently uniform, although large.

In all trials the drilling was straight, and measurements were taken, after the wheat had emerged, of the actual gaps for all plots.

AVERAGE DISTANCE BETWEEN PLOTS (INCHES)

Intended spacing (inches)	W1	I1	I2
7	6.6	6.2	7.4
14	14.0	12.0	14.5
18	18.5	16.4	18.4
28	27.8	24.2	28.3

Layout

The natural unit for this trial is the half-plot, since a spacing treatment affects one side of a plot (and one side of the adjacent plot) while the variety is sown over a whole plot. If the symbol S is used for a half-plot of the short variety when it is adjacent to a short variety plot, and S′ when it is adjacent to a tall, and if similar meanings are attached to T and T′, then the complete list of treatments is:—

S 7	S′7	T 7	T′7
S 14	S′14	T 14	T′14
S 18	S′18	T 18	T′18
S 28	S′28	T 28	T′28

As a half-plot with treatment S 7 must have another S 7 adjacent to it, each replicate contained two of each of the S and T treatments and one of each the S′ and T′. Also, as the same variety is drilled over both halves of one plot, an S treatment on one half of the plot determines that the other half must be either an S or an S′. Fig. 1 will make this clear. These facts imposed restrictions on the layout, but apart from these restrictions treatments were randomized in blocks. Five replicates were used in each trial. In the analysis of variance, "differences between adjacent plots of the same treatment" were removed from the error variance, but no other allowances were made for lack of complete randomization.

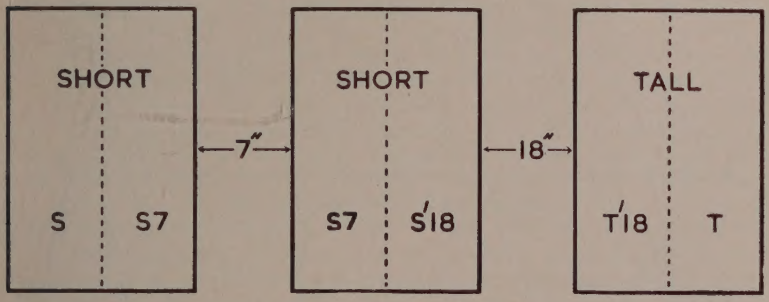

Fig. 1.—Symbols used for half plots.

Method of Harvest

In the 1952 trial the plots were two chains long. Eight sub-samples each 4 ft. in length were cut by hand from each row and bulked into one sample for each row. The middle row, which belongs to neither half-plot, was not harvested.

In the 1953 trials, continuous lengths of rows were harvested, 66 ft. for W1 and 40 ft. for I2. Each row was taken separately, including the middle row.

RESULTS

The first section deals with results from individual rows, in which the differences are often very large. The second section deals with these rows aggregated into plots, where these effects are diluted by the more stable inside rows. It is the latter figures that show more clearly the effect in routine trials of omitting the outside rows.

Individual Row Analysis

Tables 1, 2, and 3 show the yields (in bushels per acre) from each row separately, numbered from the gap in all cases, according to spacing. For rows 2 and 3 this represents actual yield, but for row 1 it represents the yield per row multiplied by the same constant as was

TABLE 1. YIELDS OF INDIVIDUAL ROWS IN BUSHELS PER ACRE IN TRIAL W1
(Based on a 7-in. width for all rows)

Spacing (inches)	Row 1	Row 2	Row 3
7	26.7	25.1	25.9
14	36.7	25.7	25.0
18	44.5	25.1	25.8
28	51.2	26.2	26.0

TABLE 2. YIELDS OF INDIVIDUAL ROWS IN BUSHELS PER ACRE IN TRIAL I1
(Based on a 7-in. width for all rows)

Spacing (inches)	Row 1	Row 2	Row 3
7	63.5	64.2	68.5
14	87.5	73.2	71.4
18	98.0	70.0	67.7
28	113.7	71.9	70.7

TABLE 3. YIELDS OF INDIVIDUAL ROWS IN BUSHELS PER ACRE IN TRIAL I2
(Based on a 7-in. width for all rows)

Spacing (inches)	Row 1	Row 2	Row 3
7	73.5	47.8	68.7
14	90.0	49.6	68.8
18	100.9	50.8	75.4
28	104.1	48.5	69.2

used for rows 2 and 3, i.e. it supposes that row 1 has a total width of 7 in. For comparisons of results, the actual yields per row are required, and it was more convenient to have them in this unit than in grammes per plot, particularly as the plot size differed in the three trials.

Table 4 shows the true yield in bushels per acre, taking into account the area available to each row.

Comparison of Spacings

Third Rows: The yields for the rows third from the outside of each plot did not show significant differences between different spacings

TABLE 4. COMPARISON OF YIELDS IN BUSHELS PER ACRE
(Based on exact area available to each row)

Spacing (inches)	Trial W1		Trial I1		Trial I2	
	Row 1	Row 3	Row 1	Row 3	Row 1	Row 3
7	27.5	25.9	68.5	68.5	71.5	68.7
14	24.5	25.0	64.3	71.4	58.6	68.8
18	24.4	25.8	59.6	67.7	55.6	75.4
28	20.6	26.0	51.4	70.7	41.3	69.2

in any of the trials; there was not even an indication of a trend. It may therefore be assumed that the effect on yield of the extra space between plots did not extend to the third row, which may consequently be taken as a standard for the centre of the plot.

Second Rows: The second row showed no significant difference between spacing treatments in trials W1 or I2. In trial I1, however, the yield of the second row at the narrow spacing (which was actually 6 in. in this trial) was significantly less than that at each of the wider spacings. It may be that the crowding effect of the 6 in. space has extended to the second row, but the evidence is rather slender.

The comparison of second row with third row showed negligible differences in trial W1 or in I1 (except at the narrow spacing already mentioned). However, in I2 there is the curious result that the second row yields were about 20 bushels below the third row *at all spacings*. It has been suggested that in a routine trial the stronger-growing outside row would depress the second row below normal, but the depression at the 7 in. spacing where no outside row effect did, in fact, exist, is as large as the others. A possible explanation for the unexpectedly low yields of row 2 would be that those coulters were set a little deeper than the others, so that the plants were retarded and then suffered a competition effect from rows 1 and 3. The field observations on this trial were that germination was even, but that at a later date the second row at the 28 in. spacing was less vigorous than other rows of those plots. All figures for the second rows in this trial are thus clearly unreliable. This should not invalidate other comparisons from this trial as rows 1 and 3 would be equally affected by an adjacent weak row. From this trial, however, there is clearly no evidence of whether a second row depression exists or not in the presence of a strong outside row. The evidence of the other two trials is that the effect of a spacing wider than 7 in. does not extend to the second row.

Outside Rows: The outside row showed marked yield increases at all spacings greater than 7 in. in all three trials, and these increases became larger with wider spacing. This may be seen clearly in Table 5, which sets out the differences between outside row and centre row at each spacing.

To summarize

Differences in yield between outside and centre rows were present in all three trials, and these increased with spacing and reached a maximum of 43 bushels, this figure being calculated on a "per row" basis.

TABLE 5. DIFFERENCES BETWEEN OUTSIDE AND CENTRE ROWS IN BUSHELS PER ACRE

(Based on a 7-in. width for all rows)

Spacing (inches)	Trial W1	Trial I1	Trial I2
7	0.3	—5.0	4.8
14	11.7**	16.1**	21.2**
18	18.7**	30.3**	25.5**
28	25.2**	43.0**	34.9**

** Differences significant at 1%.

The effect of spacing did not extend to the third row.

There is no clear evidence that difference in spacing affects the second row.

Comparison of Varieties

Table 6 shows the varietal means over all spacing for each row, and also for the increase of outside row over centre row.

TABLE 6. MEAN YIELDS OF VARIETIES IN BUSHELS PER ACRE

(Based on a 7-in. width for all rows)

Trial	Variety	Row 1	Row 2	Row 3	Row 1 - Row 3
W1	Short	40.5	26.1	26.0	14.5
	Tall	39.0	24.9	25.3	13.7
I1	Short	76.0	61.4	58.5	17.5
	Tall	105.3	78.4	80.7	24.8
I2	Short	104.1	58.7	81.9	22.2
	Tall	80.1	39.7	59.1	21.0

In W1, where the mean yields of the varieties were approximately equal, the outside row effects were also equal. The increase was 55% of the centre row yield.

In I1 the tall variety outyielded the short by 22 bushels in the centre of the plot, and also gave an outside row increase of 24.8 bushels, compared with 17.5 bushels for the short variety. However, the relative increase in each case was the same, 30% of the centre row yield.

In I2, where the short variety outyielded the tall by 23 bushels, the absolute increases in outside rows were equal for the two varieties. This increase, however, is 27% of the yield of the centre rows for the short and 36% for the tall variety.

Where varieties are compared by tests based on an analysis of variance, it is more important that the absolute difference between varieties should not be altered by a matter of technique than that their

relative difference should be so altered. However, in these three trials each of three possibilities appeared once, (a) no difference in increase, (b) an absolute difference in increase and (c) a relative difference in increase.

Competition between Varieties

What we have called the competition effect was evaluated as Σ (S-2S′) on the short variety and Σ (T-2T′) on the tall variety. It is a comparison of the yield of "short" plots adjacent to "short" plots against yield of "short" plots adjacent to "tall" plots. There were twice as many S plots as S′ plots in the trial, and this is the reason for the factor 2.

The results in the two Invermay trials did not give a clear picture. In I1 the effect "competition on the tall variety" was significant. The "outside minus centre" yields were greater by 13.6 bushels per acre for the T′ plots (tall beside short) than for the T plots (tall beside tall). The variation in this effect through the range of spacings did not follow any trend, and there was no similar effect on the short variety.

In I2 the only significant effect was the interaction "competition on the short variety × spacing". This arose because at the narrowest spacing the outside rows of the S plots (short beside short) yielded less than the outside rows of the S′ plots (short beside tall). At the other spacings the difference tended to be in the opposite direction. In this trial the short variety was the higher yielding.

In W1, where varieties yielded equally, consistent results were obtained. Yield differences were as follows, in the units of bushels per acre, based on a 7-in. width for all rows.

Outside minus centre rows for	Tall	beside	Tall	12.9 bus.
	Tall	„	„ Short	15.4 „
			Difference	2.5 „
„ „ „ „ „	Short	„	beside Tall	13.1 „
	Short	„	„ Short	15.3 „
			Difference	2.2 „

On both varieties the outside-row effect was greater by just over two bushels when the adjacent plot was short than when it was tall. This effect was not significantly affected by spacing.

Coefficients of Variation

The values of σ, the error root mean square from the analysis of variance, are given in Table 7. The outside rows were in each case much more variable than either rows 2 or 3. That this was not entirely due to the higher yields in row 1 may be seen from the coefficients of

TABLE 7. ERROR ROOT MEAN SQUARE IN GRAMMES PER ROW

	1	2	3	1-3	2-3
W1	138	69	63	111	66
I1	248	104	125	222	111
I2	249	139	192	192	181

variation in Table 8. The variate "Row 1 minus row 3" had smaller errors than the variate Row 1 above. Tests of all appropriate hypotheses were made using this variate also, but no conclusions other than those set out above were apparent.

TABLE 8. COEFFICIENT OF VARIATION

	Row		
	1	2	3
W1	14.4%	11.2%	10.2%
I1	23.5%	12.8%	15.4%
I2	18.6%	19.4%	18.7%

Comparison of 5-Row and 7-Row Plots

The comparisons of the last section have shown the effect of various spacings and competition factors on the yields of individual rows. In routine trials, it is not individual rows that are considered, but whole plots, and the effects of the variability of outside rows in such trials can be seen from consideration of half plots. Full plots cannot be used, since the layout of these trials has given different spacings on either side of each plot. The centre (i.e. 4th) row was harvested in trials W1 and I2, and consequently in these trials half plot yields were obtained from rows (1) + (2) + (3) + $\frac{1}{2}$ (4) to include the outside row and from rows (2) + (3) + $\frac{1}{2}$ (4) to exclude it. In trial I1 the centre row was not harvested and the half plots were obtained there by using (1) + (2) + $\frac{3}{2}$ (3), and (2) + $\frac{3}{2}$ (3). The arbitrary allocation of half the yield of row 4 to each of the adjacent half-plots, with no imposition of random error, is not strictly correct, but the error introduced would be so slight in comparison with total half-plot yields that this point has been ignored.

Comparison of Spacings

It is clear from Table 9 that difference in spacing had no effect on half-plot yields when the outside row was omitted, but had a marked effect when it was included.

Comparison of Varieties

Table 10 shows that the varietal differences were virtually the same whether the outside row was included or not.

TABLE 9. YIELDS OF HALF PLOTS IN BUSHELS PER ACRE FOR EACH SPACING
(Based on a 7-in. width for all rows)

	Spacing (inches)	Without Outside Row		With Outside Row	
		Row (2) + (3) + ½ (4)	Diff. from 7-in.	Rows (1) + (2) + (3) + ½ (4)	Diff. from 7-in.
W1	7	24.9		25.4	
	14	24.7	0.2	28.2	2.8**
	18	24.7	-0.2	30.3	4.9**
	28	25.3	0.4	32.7	7.3**
Sig. diff. 5%*			N.S.		1.5
1%**			—		2.0
I1	7	66.8		65.8	
	14	72.1	5.3	76.4	10.6**
	18	68.7	1.9	77.0	11.2**
	28	71.2	4.4	83.4	17.6**
Sig. diff. 5%*			N.S.		5.6
1%**					7.4
I2	7	59.8	—	65.2	
	14	60.8	1.0	69.1	3.9
	18	64.7	4.9	75.0	9.8**
	28	60.7	0.9	73.1	7.9**
Sig. diff. 5%*			N.S.		5.7
1%**					7.5

TABLE 10. YIELDS OF HALF PLOTS IN BUSHELS PER ACRE FOR EACH VARIETY
(Based on a 7-in. width for all rows)

		Without Outside Row		With Outside Row	
	Variety	(2) + (3) + ½ (4)	Diff.	(1) + (2) + (3) + ½ (4)	Diff.
W1	Short	25.4	1.0*	29.7	1.1
	Tall	24.4		28.6	
I1	Short	59.6	20.2**	64.2	22.9**
	Tall	79.8		87.1	
I2	Short	72.1	21.2**	81.7	22.0**
	Tall	50.9		59.7	

Competition between Varieties

In neither the 5- nor the 7-rowed plots was the competition effect significant.

Coefficients of Variation

In theory, the coefficient of variation is expected to decrease as the plot size is increased. Table 11 shows, however, that in these trials the coefficient of variation is little different whether outside rows are included or not, the reason being that the outside row is more variable

TABLE 11. COEFFICIENTS OF VARIATION OF HALF PLOTS

	Without Outside Row (2) + (3) + ½ (4)	With Outside Row (1) + (2) + (3) + ½ (4)
W1	9.7%	10.6%
I1	12.7%	14.1%
I2	16.2%	16.4%

than the others. These C.V's have been computed, eliminating from the error the variance due to the spacings means, and they represent the case where the drilling is very accurate and the spacing between plots is always as intended. In the 7-row plots, i.e. including the outside rows, the pooled variances used in Table 11 conceal some differences and the separate C.V.'s for each spacing are shown in Table 12. (Spacing differences do not affect the errors in 5-row plots, so C.V's from pooled variances are there satisfactory). It can be seen that at the 7-in. spacing the C.V. for the 7-row plots is always higher than the corresponding C.V. for the 5-row plots. Although there is no significant "outside row effect" in terms of mean yield at this spacing, probably small deviations from the exact spacing here exercise a marked effect.

TABLE 12. COEFFICIENTS OF VARIATION OF HALF PLOTS AT SEPARATE SPACINGS, WITH OUTSIDE ROWS

Spacing (inches)	Trial W1	Trial I1	Trial I2
7	11.2%	15.1%	25.7%
14	8.4%	10.9%	19.2%
18	11.5%	10.9%	8.1%
28	12.8%	17.2%	16.1%
5 row plots, all	9.7%	12.7%	16.2%

In the plots at 14 in. and 18 in., the C.V's for 7 rows vary about the C.V's for the corresponding 5 rows and would seem to give an equal average effect. At 28-in. spacings the C.V's for the 7-row plots are either greater than or equal to the corresponding 5-row C.V.

In order to estimate the effect of irregular drilling in a routine trial, the coefficients of variation were obtained from an analysis of the plots in the 7, 14, and 18-in. spacings only, and the component of variation due to spacings was then not removed from the error variance. That is, it is assumed that the spacing can vary from 7 to 18 in., which is not unlikely in a poor trial. These recomputed values of the coefficients of variation are shown in Table 13, where it is clear that the exclusion of outside rows has given a worthwhile reduction in error for each trial. The magnitude of the errors is about twice as high as would be considered satisfactory in a standard trial, but these errors were based on plots of half width and the length of row harvested was only about one quarter of that taken in a standard trial.

TABLE 13. COEFFICIENTS OF VARIATION OMITTING 28-IN. ROWS AND IGNORING SPACING DIFFERENCES

	Without Outside Row (2) + (3) + ½ (4)	With Outside Row (1) + (2) + (3) + ½ (4)
W1	9.5%	13.6%
I1	12.3%	15.4%
I2	16.7%	18.3%

DISCUSSION

The questions raised in the introduction may now be answered as far as the evidence from these trials allows.

(a) The outside row effect is very sensitive to slight differences in spacing between plots. This applies whether individual rows or half plots are considered. Thus, unless the drilling of a trial is very accurate, individual plot yields are liable to have high errors if outside rows are not discarded.

(b) There is no evidence that inclusion of the outside rows affects the estimates of the relative performance of varieties.

(c) There is some evidence of a consistent competition effect in one trial and a strong but inexplicable effect in another trial when individual rows are considered, but none when half-plots are considered.

(d) On an average of all spacings, the coefficient of variation is little affected by the outside row effect if the spacing is very regular. No one particular spacing emerged from the 3 trials as being particularly good (from a variance point of view). The 7-in. and 28-in. spacings showed for 7-row plots errors that were rather higher than the average for 5-row plots, which was constant over all spacings. If, however, spacings irregularities between 7 and 18 in. are counted as error components, there is a consistent reduction in C.V. if outside rows are eliminated.

These answers must be discussed in relation to the practical advantages and disadvantages of various techniques. These results can be considered applicable to varietal trials only and not to manurial trials where actual transfer of nutrients may occur as well as competition between plots. They are also limited to variety trials where varieties do not differ very greatly in height or yield ability.

Inclusion of the outside rows always increases the estimate of yield at all spacings greater than 7 in., unless allowance is made for part of the gap between plots to be included in the plot size. For comparative purposes this upward bias in all yields may not be important provided it is constant over all varieties. When this becomes great, as for instance sometimes at the 28-in. spacings, there may be difficulty in assessing the practical value of treatment differences which should be always judged against the general level of yields of the trial. These trials have shown that where drilling is regular, there is no consistent favouring of either the taller or the higher yielding variety, the effect being different in all 3 trials.

However, since the yield of outside rows is increased by almost 2 bushels per acre for every extra inch of gap width, irregularities in spacing could markedly affect means. Evidence of the effect of competition between varieties was unfortunately not clear cut. It appeared in individual row comparisons but not on a plot basis. If the effect can sometimes occur it would be unwise to adopt a technique which would encourage it, although till evidence on the probability of occurrence of the effect is obtained, this point alone would not justify the exclusion of the outside rows.

Although it might seem that since no outside row effect was shown to exist at the 7-in. spacing, this might be the ideal distance if it were desired to harvest whole plots, in fact the variability of 7-row plots at 7-in. spacing was greater than the variability of 5-row plots at any spacing. The practical difficulties of sowing, observing and harvesting a trial at this spacing can be imagined, and were instanced in trial I2 where, although much care was taken at sowing, the plots in two instances ran together for part of their length, so that the separate yields for the respective outside rows could not be obtained. Thus the 7-in. spacing can be discounted for routine trials.

At the 28-in. spacing, the design of these trials did not allow the effect of small irregularities in drilling to be measured. However as the coefficients of variation for 7-row plots at this spacing were larger than those for 5-row plots, no advantage is gained by using this spacing. Against this is the marked practical disadvantages of increased weed growth and a gross over-estimate of the yield.

At the 14- and 18-in. spacings, which are the most convenient practically and have now been shown to be at least not worse than the wider or narrower spacings as regards bias and variance, no reduction in variance can be expected from elimination of the outside rows if drilling is accurate to within 2 in. The possibility of a competition effect must be considered in making the decision to harvest all rows in a well drilled trial. Where the drilling is more irregular, a consistent reduction in variance is achieved by eliminating outside rows. This, although small, is important, since in variety trials it is desirable to be able to detect small differences, yet in many cases the amount of seed available is limited, so that the variance of means cannot be reduced by increasing plot size or replications.

The main conclusions to be drawn from these trials are:

(1) That no departure from 14-in. spacings is necessary although 18 in. would be almost equally satisfactory.

(2) That outside rows should be removed if the spaces between plots in a trial are uneven, as this will give a worthwhile reduction in error variance.

(3) That where drilling is so accurate that spaces between plots do not vary by more than about 2 in., the error variance will not be reduced by omitting the outside rows.

(4) The competition effects are not clear-cut, and although it seems unlikely that they will be large, the possibility of their existence may provide sufficient reason for omitting outside rows in some accurately spaced trials.

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A NOTE ON THE EFFICIENCY OF *Bombus terrestris* (L.) AS A POLLINATOR OF LUCERNE (*Medicago sativa* L.)

By L. GURR, Massey Agricultural College, Palmerston North*

(Received for publication, 18 August 1955)

At Omaka, Marlborough, in February 1954, although honey-bees and *B. ruderatus* (Fab.) were freely working lucerne (*Medicago sativa* L.) for nectar, none was seen to trip the flowers. They preferred to alight on the side of the flower and insert their tongues between the overlapping parts of the standard and wing petals: thus they obtained the nectar without being showered with pollen, and consequently did not effect pollination. *B. terrestris* (L.), however, invariably tripped the flower even when collecting nectar. Since these observations were made, Brian (1954) has reported similar findings by two groups of European workers (Akerberg and Lesins in Central Sweden, and Stapel in Denmark). They found that *B. terrestris* tripped 78% of lucerne flowers visited, whereas only 13% were tripped by *B. agrorum* (a long-tongued bumble bee like *B. ruderatus*).

When an insect trips a lucerne flower, the released stigma and stamens strike its body dusting it with pollen. If the insect has previously visited a flower of another plant, pollen from this is probably transferred to the receptive stigma and thus cross-pollination takes place. Only a very small proportion (1.6% according to Hadfield and Calder, 1936) of untripped lucerne florets produce seed. Hadfield and Calder (*ibid.*) showed that cross-fertilization resulted in a marked increase in seed production whereas self pollination produced the reverse effect so that cross-fertilization, which is only produced by insect pollination, is necessary for good seed-set.

Bombus terrestris (L.) when visiting red clover (*Trifolium pratense* L.) flowers sometimes bites holes in the corolla tube close to the nectary, thereby obtaining nectar supplies without effecting pollination. This habit, although not resorted to by all nectar-gathering individuals and of course not by pollen gatherers of this species, has led to *B. terrestris* being condemned by growers of both clover and lucerne seed alike, as an unfortunate introduction to this country. This view is, however, shown to be incorrect for, although *B. ruderatus* is undoubtedly more efficient than *B. terrestris* as a pollinator of red clover, *B. terrestris* is the more useful species in the pollination of lucerne, because it trips more lucerne flowers than *B. ruderatus*, and so is a more valuable species than was hitherto recognized.

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* The information presented in this note was gained while the author was a member of the staff of the Entomological Research Station, Nelson, N.Z.

PASTURE QUALITY AND RUMINANT DIGESTION

I. SEASONAL CHANGE IN BOTANICAL AND CHEMICAL COMPOSITION OF PASTURE

By A. T. Johns, Grasslands Division, Department of Scientific and Industrial Research, Palmerston North, New Zealand

(Received for publication, 4 May 1955)

Summary

The seasonal change in botanical and chemical composition of a pasture of short-rotation ryegrass and clover was determined at frequent intervals over a 17-month period. Values for total N, non-protein N, free and bound amino N, and soluble carbohydrate are presented.

The standard errors of the sampling procedure and chemical estimations have been determined.

The most striking change is the rapid fall of protein level in the late spring and rapid rise in the autumn.

Introduction

New Zealand farmers rely almost solely on pasture herbage, of which only a small amount is fed as hay or silage, as food for sheep and cattle. The adequacy of pasture herbage as the sole diet of domestic animals and the efficiency with which it is converted into milk, meat, and wool, are therefore major problems in the livestock industry.

A number of metabolic disorders of ruminants occur in New Zealand. In late winter and early spring, trouble is experienced with grass staggers (hypomagnesaemia), ketosis, milk fever (hypocalcaemia), and sleepy sickness (pregnancy toxaemia); in spring and early summer, bloat is a serious problem; in autumn, facial eczema and lack of thrift in hoggets sometimes result in major stock losses. Considerable research effort has been applied to all these disorders and it is increasingly apparent that the classical feed and forage analyses are inadequate for an understanding of their aetiology and hence for the application of control measures. A much more detailed knowledge is required of the chemical composition of the pasture herbage, of the behaviour of individual metabolites when subjected to ruminant fermentation, and of the subsequent metabolism by the animal of the end products of ruminant digestion. The non-structural constituents, consisting of soluble materials of low molecular weight and of protein, are of particular significance, since they are likely to be the most reactive compounds in the rumen. Ferguson (1948) has shown that these constituents may amount to over 55% of the dry herbage, while Bathurst (pers. comm.) has found values of 45% to 55%.

In the present study, which deals with limited aspects of a larger investigation into pasture quality, particular attention is paid to changes in level in nitrogenous compounds. These were chosen for study because of the high protein levels that are characteristic of the most productive pastures and that are considerably in excess of generally accepted production and maintenance standards for both sheep and cattle. The ecology of such pastures has been intensively studied by Sears (1953), whose work provides an explanation of herbage nitrogen levels largely in terms of the effect of clovers and the grazing animal.

A number of investigators have been concerned with the effect of high protein intake on the health and production of ruminants. Sjollema (1950) believes that high protein and potassium levels may be predisposing factors in the production of grass tetany. He also states that protein-rich grass has an adverse effect on milk production and possibly on the health of the animal. He advocates the use of supplementary feeding with protein-poor ratios during the period when the protein content of the grass is high.

White (1953) states that high-protein low-carbohydrate diets may be a predisposing factor in a number of nutritional and metabolic disorders of ruminants such as ketosis and hypomagnesaemia. King (1952) believes that unrestricted grazing of young spring grass of low fibre and high protein content would have a depressing effect upon the fat percentage of milk if the ration were not supplemented to maintain a reasonable fibre level.

In ruminants, part of the food protein is broken down by micro-organisms and the remainder passes on to the abomasum. Of the protein attacked by the rumen flora, part is assimilated by the micro-organisms, which also pass on to the true stomach, and part is converted to ammonia which may be absorbed by the animal directly from the rumen (McDonald, 1948). The non-protein nitrogenous fractions of the food are dealt with in a similar manner. As Synge (1952) has pointed out, "It is evident that quite other factors than amino acid composition determine the value of protein or non-protein nitrogen to a ruminant animal: the amount and nature of the carbohydrate that is being simultaneously fermented, the solubility of the protein and its susceptibility to bacterial proteases are obviously among the important factors."

If high-protein low-carbohydrate diets are deleterious to animal health it is likely that the effect may be mediated by the products of digestion in the rumen. The present paper deals with changes in levels of certain nitrogenous and soluble carbohydrate fractions of high-production pasture during a period of nearly two years; in Part II the levels of volatile fatty acids and ammonia in the rumen of sheep grazing the same herbage are given.

Methods

An area of $\frac{1}{10}$ acre was sown in pasture on 29 March 1952 with a mixture of 1.5 lb. short-rotation ryegrass, 0.3 lb. pedigree white clover, and 0.4 lb. of red clover. Before sampling started in November 1952, the area had been grazed with sheep at approximately monthly intervals.

During the experimental period, enough animals were placed on the area to eat it down within a period of three days.

Sampling Procedure

During the preliminary period, November–December 1952, two separate samples were taken at weekly intervals. Each sample was obtained by dividing the area into 16 sub-areas and cutting a foot square randomly chosen in each, care being taken that successive samplings did not fall on the same sites. After the preliminary samplings to determine the error involved in the technique, a single sample of 16 one-foot squares similarly located was taken at each date.

Samples were taken at 9 a.m. A representative portion of the sample (400 to 500 g.) was taken and dissected into grasses and clovers. These were immediately freeze dried (Bathurst and Allison 1949), ground, and stored in tightly stoppered jars. The residual water content was approximately 5% for all samples.

Extraction Procedure

Dried material (2 g.) was treated in a Waring blendor with 4 portions of 100 ml. of 80% alcohol. Each blending occupied 3 min. The combined extracts were concentrated under reduced pressure until all the alcohol had been removed, the residue being made up to 10 ml. with water.

The plant residue after extraction was blended with 4 portions of 100 ml. of water. The combined water extracts were concentrated under reduced pressure and made up to 10 ml.

Nitrogen. Total nitrogen was determined by macro-kjeldahl using 0.5 g. of dried tissue. Nitrogen of the extracts was determined by the micro-method of Reifer (1940). The distillation was carried out according to procedure of Ma and Zuazaga (1942) in the Markham steam-distillation apparatus. The ammonia was collected in boric acid solution and titrated with 0.01 N standard acid.

Sugars. Total reducing sugars in the alcohol and water extracts were determined after hydrolysis with H_2SO_4 and clearing by the method of Doak (1939). The sugars were estimated by the ferricyanide method of Fujita and Iwatake (1932). The carbohydrate results are reported in terms of glucose.

Amino acids were determined by the quantitative paper chromatographic method of Thompson *et al.* (1951).

RESULTS

Sampling Errors

The results of the samplings between 11 November and 16 December 1952 are shown in Table 1.

The field was grazed with sheep on 28 November after the first series of samples.

A sample collected on 21 November 1952 consisted of ryegrass 69%, *Poa* spp. 1%, red clover 29%, and white clover 3%, (dry-weight basis), while on 16 December 1952 ryegrass contributed 60%, *Poa* spp. 2%, red clover 24%, and white clover 17%.

Omitting the first date (on which occasion two observers took separate samples), S. E. of mean percentage at any date was $\pm$ 1.70 and the error in any one sample (16 sq. ft.) was $\pm$ 2.9.

These figures include both sampling variance in the field and chemical determination variance.

The standard errors for the extraction procedure and chemical determination were found by carrying out 4 separate extractions and sets of chemical determinations on 3 separate ryegrass samples.

TABLE 1. CHANGE IN CHEMICAL COMPOSITION OF GRASS AND CLOVER WITH STAGE OF GROWTH

(Means of two samples for each date)

Date	Height of Pasture in.	Green Sample Weight lb.	Species	D.M. %	Species %	Total N %	N.P.N. Alcohol Extract mg. %	N.P.N. Water Extract mg. %	Total N.P.N. mg. %	Total Soluble Sugars % of D.M.	Total N.P.N. as % of N
11.11.52	6	—	Clover	17.6	42.3	5.24	800	150	950	3.1	17.7
			Grass	17.0	57.6	4.54	860	92	952	4.2	22.5
18.11.52	9	3.2	Clover	13.7	39.5	4.68	545	99	644	3.9	13
			Grass	14.3	60.5	4.16	565	49	614	8.2	14.8
24.11.52	12–15	6.9	Clover	12.1	35	4.52	614	103	717	4.5	15.7
			Grass	13.6	65	3.74	667	114	781	6.6	19.7
Grazed 28th November. Regrowth											
1.12.52	3–5	2.1	Clover	16.5	28	3.60	463	68	531	5.5	14.4
			Grass	16.9	72	3.02	458	89	547	5.6	18.5
12.12.52	9	3.5	Clover	16.3	35	3.93					
			Grass	16.8	65	3.19					
16.12.52	12–15	4.8	Clover	12.5	40	3.59					
			Grass	13.9	60	3.06					

STANDARD ERRORS PER SAMPLE (16 sq. ft.)

Species	Species %	N.P.N. Alcohol Extract mg. %	N.P.N. Water Extract mg. %	Total N %	Total Soluble Sugars %
Clover	± 2.9	± 91	± 6.8	± 0.25	± 0.55
Grass	± 2.9	± 54	± 11.9	± 0.12	± 1.27
Both		± 75	± 9.7	± 0.19	± 0.98

The contribution of the extraction procedure and chemical determinations on the grass to the overall standard error, as found from the 12 extractions, is as follows:

Alcohol extract N.	± 15.0 mg. %	sugar ± 0.45 %
Water extract N.	± 8.6 mg. %	sugar ± 0.83 %
Total N.	± 0.03 %	

Chemical Composition of Pasture at Various Heights Above Ground

A sample of short-rotation ryegrass 12 in. tall was taken from the field and cut into 3 lengths of 4 in. to determine differences in selected

constituents at various heights from the ground. The results are given in Table 2.

TABLE 2. CONCENTRATION OF CONSTITUENTS OF RYEGRASS AT DIFFERENT HEIGHTS ABOVE GROUND LEVEL

Height (in.)	D.M. %	% of Total D.M.	Total N %	N.P.N. Alcohol Extract mg. %	N.P.N. Water Extract mg. %	Total N.P.N. mg. %	Sugar Alcohol Extract % D.M	Sugar Water Extract % D.M	Total Sugar %
0– 4	13.0	40.4	2.80	252	172	424	3.2	0.9	4.1
4– 8	14.5	39.6	3.79	228	151	379	4.5	0.3	4.8
8–12	18.0	20.0	4.31	224	99	343	3.8	0.4	4.2

Analyses for the constituent amino acids of the alcohol extract are shown in Table 3.

TABLE 3. AMINO ACID CONTENT (mg. %) OF ALCOHOL EXTRACT OF SAMPLES OF RYEGRASS AT DIFFERENT HEIGHTS ABOVE GROUND

Amino Acid	0 – 4 in.	4 – 8 in.	8 – 12 in.
Aspartic acid	160	210	270
Glutamic acid	180	265	375
Asparagine	60	60	Trace
Glutamine	110	50	50
Alanine	20	20	25
Serine	45	40	20
γ amino butyric acid	365	320	210
Threonine	Trace	Trace	Trace

The error of the determination of the amino acids is of the order ± 10% except for γ amino butyric which is ± 20%.

Seasonal Changes in Chemical Composition of the Grasses and Clovers of a Grazed Pasture

The results of samplings are given in Table 4. The intervals between samplings were not regular nor were samplings made at the same height of pasture. When clover formed a very small part of the herbage, clover analyses were omitted.

Amino acids were determined on all alcohol extracts but as no consistent trends could be found the ranges only are given in Table 5.

TABLE 4. SEASONAL CHANGE IN BOTANICAL AND CHEMICAL

Date	Species %	Grass D.M. %	Total Nitrogen %	N.P.N. Alcohol Extract mg. %	N.P.N. Water Extract mg. %	Total N.P.N. mg. %	N.P.N. % of Total N	Sugar Alcohol Extract % D.M.	Sugar Water Extract % D.M.	Total Sugar % D.M.
				Grass						
23.3.53	C 30 G 70	19.0	3.14	236	101	337	10.7			
13.4.53	C 20 G 80	16.0	3.84	248	118	366	9.5	0.9	0.7	1.6
4.6.53	C 6 G 94	14.0	4.60	300	291	591	12.8	4.3	0.7	5.0
22.6.53	C 2 G 98	11.5	4.70	280	133	413	8.8	6.7	0.8	7.5
17.7.53	C 2 G 98	15.0	4.34	360	144	504	11.6	3.5	0.2	3.7
17.8.53	C 0 G100	13.4	5.49	585	79	664	12.1	3.6	0.2	3.8
15.9.53	C 3 G 97	12.0	5.19	420	77	497	9.6	4.0	0.4	4.4
12.10.53	C 4 G 96	14.0	4.30	316	101	417	9.7	6.0	0.5	6.5
23.10.53	C 2 G 96	15.0	4.15	460	72	532	12.8	3.9	0.4	4.3
12.11.53	C 19 G 81	19.5	3.18	420	62	482	15.1	10.0	1.2	11.2
22.12.53	C 23 G 77	16.0	3.20	232	74	306	9.6	1.8	0.2	2.0
11.1.54	C 48 G 52	13.0	3.20	213	52	265	8.3	5.1	0.9	6.0
11.2.54	C 37 G 63	20.0	3.12	175	41	216	6.9	4.2	0.2	4.4
5.3.54	C 44 G 56	32.0	2.44	266	129	395	16.2	2.2	0.1	2.3
2.4.54	C 70 G 30	25.0	4.10	280	33	313	7.8	5.8	0.4	6.2
4.5.54	C 53 G 47	17.0	4.40	315	46	361	7.9	6.1	0.2	6.3
16.7.54	C 21 G 79	21.9	4.20	220	119	339	8.1	4.1	0.5	4.6
6.8.54	C 12 G 88	17.5	5.07	546	95	641	12.6	2.4	0	2.4

COMPOSITION OF RYECRASS—CLOVER PASTURE

Clover									
Clover D.M. %	Total N %	N.P.N. Alcohol Extract mg. %	N.P.N. Water Extract mg. %	Total N.P.N. mg. %	N.P.N. % of Total N	Sugar Alcohol Extract % D.M.	Sugar Water Extract % D.M.	Total Sugar % D.M.	Height at Grazing in.
19.0	4.57	409	92	511	11.2				
15.7	4.63	332	139	471	10.2	5.2	6.2	11.4	6
12.2	4.80	291	291	582	12.1	3.6	0.4	4.0	4–10
9.0	4.20								2–4
13.0	4.14								6
									6–8
11.5	5.06								6–9
13.0	4.31								4–12
13.0	4.85								4
16.0	4.38	390	60	450	10.3	9.7	0.4	10.1	6–12
13.0	4.31	210	51	261	6.1	0.7	0.3	1.0	4–7
12.0	4.4	150	72	222	5.0	1.3	0.6	1.9	4–8
21.0	4.57	175	70	245	5.4	4.6	0.5	5.1	6
27.0	4.74	350	88	438	9.3	3.1	0.4	3.5	3
20.0	4.95	318	21	339	6.9	4.3	0.3	4.6	6–8
16.6	5.34	350	178	528	9.9	3.7	0.5	4.2	6–8
17.6	4.38	463	220	683	15.6	2.3	0.7	3.0	6–8
15.7	5.14	634	174	808	15.7	2.5	0.2	2.7	8

TABLE 5. RANGE IN CONCENTRATION OF AMINO ACIDS IN 80% ALCOHOL EXTRACTS OF GRASS AND CLOVER (mg. %)

Amino Acid	Grass	Clover
Aspartic acid	50 – 280	50 – 210
Glutamic acid	60 – 635	70 – 320
Asparagine	Trace – 440	40 –1240
Glutamine	Trace – 220	Trace – 90
Alanine	Trace – 210	Trace – 115
Serine	Trace – 75	Trace – 70
γ amino butyric acid	Trace – 250	Trace – 140
Threonine	Trace – 70	Trace – 50

DISCUSSION

No solution has been found of the problem of sampling a pasture to simulate grazing. In the present series of experiments it was considered of primary importance that the animals graze normally, and the method of sampling by means of randomly distributed square-foot plots was therefore adopted. This procedure is recognized as being a compromise solution, since it gives an approximation to the food actually ingested by the animal. Nevertheless, the low standard errors show that considerable confidence can be placed in the method as a means of determining the feed available to the animal. Botanical composition changed greatly during the experiment, from complete grass dominance between June and October 1953, to marked clover dominance in April 1954.

The primary purpose of the experiment was a study of nitrogenous compounds and sugars through a complete annual grazing cycle. As has been demonstrated previously (Melville and Sears 1953), total nitrogen of the grass component shows a marked seasonal fluctuation, being high in late autumn, winter, and early spring, and low in summer and early autumn. The fluctuation in grass nitrogen levels from 2.4% to 5.5% is offset by the relative constancy of the clover nitrogen (4.1% to 5.3%) and by marked clover dominance in the association during summer and autumn. As a result the herbage nitrogen varies only between 3.4% and 5.5%.

Non-protein nitrogen, calculated as the nitrogen extracted by 80% alcohol followed by water, varies between 5% and 22% of the total nitrogen over the period covered by Tables 1 and 4. There is no consistent variation in percentage due to species, to stage of growth, or to total nitrogen level. These findings derived from a pasture under grazing are consistent with previously published values (Bathurst 1953; Bathurst and Allison, 1949).

The amino acid analyses performed on the alcohol extracts are not presented in full since large variations occurred which bore no

relation to season (Table 5). From this and other unpublished studies (Bathurst, pers. comm.) it is apparent that factors governing concentration of individual amino acids are still quite unknown. Table 5 confirms, however, that the free amino-N of pasture herbage is in the form of glutamine, asparagine, serine, alanine, and aspartic, glutamic, and γ amino butyric acids.

From Bathurst's (1953) figures it may be assumed that the contribution by free amino acids to alcohol-soluble nitrogen ranges from 50% to 80%, while the water-soluble nitrogen exists very largely in the form of bound amino acids. It is obvious therefore that well over 90% of the total organic nitrogen ingested by the animal is derived from amino acids in either free or bound form. The remainder, which comprises free purines, N-methylated compounds such as choline and betaines, and unknown compounds, is obviously of small importance quantitatively; only for materials with high pharmacological activity would the non-amino-acid fraction assume importance in nutritional studies.

The partition of the total amino acids among free, bound (water soluble), and bound (water insoluble), may however be of considerable importance in determining the pattern of ruminant fermentation over the early digestion period. The first two categories would represent the most available nitrogenous constituents in the herbage, and the results of Sirotnak *et al.* (1953) show that different amino acids lead to different proportions of volatile fatty acids.

The chief points of interest in the soluble sugar values are their relative constancy throughout the year and the low absolute values compared with those quoted by Waite and Boyd (1953) and by Laidlaw and Reid (1952). At no season and in no sample analysed did the total soluble sugars approach those quoted by these investigators who report values up to 26.6% as compared with the maximum value of 11.2% found in this investigation. The difference probably lies in the fructosan fraction which under Scottish conditions reaches 16.6% of the dry tissue. A further cause of the discrepancy may lie in the fact that all samples in this investigation were taken at 9 a.m. Diurnal variations in soluble carbohydrate levels are such that, had samples been taken after midday, considerably higher values would probably have been recorded.

It should be emphasized that, in contrast to the fairly complete picture emerging from the nitrogenous constituents, the carbohydrate analyses give only a portion of the fermentable non-nitrogenous constituents of the protoplast. Unfortunately organic acid analyses have proved difficult to place on a routine basis and they have been excluded, but evidence, largely unpublished (cf. Mitchell, 1953), indicates that even with their inclusion this fraction is still incompletely defined.

The difference in chemical composition in pasture herbage at varying heights above ground level has long been reognized (Fagan, 1928; Waite and Boyd, 1953). Tables 2 and 3 give more precise information than has been hitherto available, in that the nitrogenous constituents have been analysed in detail. A clearer picture of amino acid variation emerges from vertical partition of the herbage than from

the seasonal analyses, with the dicarboxylic acids higher in the flag than in the sheath, and the reverse being true for glutamine, asparagine, and γ amino butyric acid. The results emphasize the difficulty encountered in sampling to simulate grazing. At any one time grazing animals may have available to them the leaf flag, the whole shoot, and the leaf sheath, with considerable differences in the composition of the ingested feed.

The data also provide interesting information on the flexibility of grazing possible with short-rotation ryegrass, whose characteristics are high winter production and high palatability at heights up to 18 in. Table 1 demonstrates that crude protein declines relatively little between the 6- and 15-in. stages of growth. Indeed in the samplings in November 1952 protein content is only slightly lower at the 12- to 15-in. stage than 12 days earlier at the 3- to 5-in. stage. At both dates the grass protein levels are above the standards laid down for livestock maintenance and production, without any assistance from the clover component.

It is clear that the statement by Watson (1952) that "the value of herbage as regards its energy and protein value is correlated with its age, which in turn is related to the leafiness of the sward" must be modified under New Zealand conditions to take into account the season of the year. The analyses for clover and grass in Table 4 show that, with the bred strains of grasses and clovers available in New Zealand, pastures can maintain a high protein and low soluble-carbohydrate level throughout the year.

The protein level is evidently in excess of animal requirements (Sjollema, 1950) and may be considered as waste. However, a large proportion of this protein is returned to the sward in the form of dung and urine and results in the maintenance of high pasture yields.

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FACTORS AFFECTING INCIDENCE OF SUPERFICIAL SCALD IN THE ROME BEAUTY APPLE AND ITS COLOURED SPORT, FRIMLEY BEAUTY

By C. A. S. Padfield, Fruit Research Station, Department of Scientific and Industrial Research, Havelock North

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Summary

Development of superficial scald on Frimley Beauty and Rome Beauty apples was studied over a period of four years in relation to time of picking, delay between harvest and cool storage, and the use of oiled wrappers.

There were indications that early picking (late March) favoured scald development. Delay between picking and cool storage in general caused a marked increase in the incidence of scald.

Oiled wraps did not eliminate scald, but their use consistently reduced the percentage loss. The use of oiled wraps for both these varieties is recommended.

Introduction

In most years, superficial scald appears on the apple variety, Rome Beauty, and its more highly coloured sport, Frimley Beauty, in cool store. This disorder appears to be particularly troublesome on fruit grown in Hawke's Bay, where in some seasons a large proportion of the commercial stocks is affected.

An examination of the records of receipts at cool stores for 1950 showed that harvesting of both varieties was spread over considerable periods: Rome Beauty from the first week of April until the second week of June; Frimley Beauty from the first week in April to the last in May.

It is known too that there is often a considerable delay between harvest and the time the fruit is cool stored.

It is not standard practice in New Zealand to wrap these varieties in oiled paper.

Experiments were undertaken to assess the importance of time of picking and delay in cool storage and the effect of oiled wrappers on scald incidence in these varieties.

Experimental

Fruit of these two varieties was obtained from seven orchards over the years 1951–54; Frimley Beauty apples were obtained from orchards 1 to 4, while Rome Beauty came from orchards 5 to 7. Sample size varied between experiments from 200 to 600 apples per treatment.

Effect of Maturity and of Oiled Wrappers

In 1951, trial samples of both varieties were taken at random from commercial picks made at the various orchards.

Samples comprised four bushel cases of fruit in plain wraps and four in oiled wraps. It was intended that samples should be obtained from each orchard at two harvesting dates and that fruit should be placed

in cool store as quickly as possible after harvest. It will be seen that there was some deviation from this procedure for a number of samples (Tables 1, 2). Immediately on removal from cool store, one case of each sample was examined for scald, but the remaining three cases were not examined until the fruit had been held for a further 14 days at air temperature. It was not discovered until after examination that fruit from Orchard 2 was not submitted until 26 days after harvesting.

In 1952 a similar experiment was carried out. Results from the second examination in both years are given in Table 1.

TABLE 1. EFFECT OF ORCHARD, PICKING DATE, DELAY IN STORAGE, AND TYPE OF WRAP. 1951, 1952

Orchard	Picking Date	Delay between Harvest and Storage (days)	Period between Harvest and Post-storage Examination (days)	Percentage Scald	
				Plain Wraps	Oiled Wraps
Frimley Beauty 1951					
1	17 April	6	112	68	31
	8 May	6	91	4	0
2	22 April	26	107	18	8
Frimley Beauty 1952					
1	22 April	2	126	16	3
3	5 May	3	98	30	17
	19 May	2	83	14	5
4	29 April	2	118	3	3
Rome Beauty 1951					
5	5 May	9	93	0	0
6	25 April	5	103	0	0
	9 May	2	89	0	0
7	10 May	4	88	0	0
Rome Beauty 1952					
5	8 April	1	140	1	0
	22 April	1	126	2	0
6	16 April	1	134	1	0
	29 April	2	121	8	4
7	16 April	2	134	43	27
	6 May	6	113	22	9

No scald was apparent on the fruit examined immediately after removal from store in either year.

Effect of Time of Picking

As both these varieties are harvested commercially over a long period, it was of interest to ascertain whether the time of picking had any influence on subsequent incidence of scald. Successive picks were made on two orchards for each variety, starting on 31 March and at approximately weekly intervals thereafter until the trees were stripped at the last commercial pick.

Samples consisted of two bushels at each picking time taken from eight trees, the same trees being used at each harvesting date. All fruit

TABLE 2. EFFECT OF ORCHARD AND PICKING DATE 1952

Orchard No.	Picking and Storage Date	Period between Harvest and Examination (days)	Scald %
Frimley Beauty 1952			
1	31 March	148	58
	7 April	141	52
	15 April	133	40
	22 April	126	60
3	31 March	129	54
	7 April	122	19
	15 April	114	9
	21 April	108	24
	7 May	92	15
	12 May	87	14
	19 May	80	14
Rome Beauty 1952			
5	31 March	148	1
	7 April	141	1
	15 April	133	0
	22 April	126	0
7	31 March	149	44
	7 April	142	11
	15 April	134	15
	21 April	128	15
	6 May	113	23
	12 May	107	10

was wrapped in plain sulphite paper and placed in cool store on the day of harvesting. Samples were examined immediately on removal from store and again after being held for 14 days at air temperature.

Results after 14 days are given in Table 2.

Effect of Delay between Harvest and Cool Storage

Nine bushels of fruit packed in plain sulphite wraps were obtained at each harvesting date in 1953. These were sub-divided into lots of 3, the first being placed in store within 24 hours of harvesting, followed by three after 7 days, and the remainder after 14 days. During this interval

TABLE 3. EFFECT OF ORCHARD, PICKING DATE, AND DELAY IN STORAGE 1953

Orchard No.	Picking Date	Delay between Harvest and Storage (days)	Period between Harvest and Post-storage Examination (days)	Scald %
Frimley Beauty				
1	20 April	1	71	7
		7	71	33
		14	71	39
4	19 April	1	72	1
		7	72	17
		14	72	76
4	7 May	1	64	0
		7	64	0
		14	64	0
Rome Beauty				
5	16 April	1	104	2
		7	104	3
		14	104	75
6	16 April	1	103	1
		7	103	1
		14	103	19
7	13 April	1	105	1
		8	105	1
		14	105	3
7	22 April	1	97	3
		7	97	6
		14	97	33

the fruit was held at air temperature. Fruit was removed from store when the first signs of scald appeared, and held for a period of 14 days at air temperature before examination.

Results are given in Table 3.

Effect of Picking Date, Oiled Wraps, and Delay between Harvest and Storage

In previous years the effects of oiled wraps and of delay between harvest and storage on scald development had been investigated. In 1954 the effect of a combination of oiled wraps and delay between harvest and storage on scald incidence was examined, using the variety Rome Beauty from Orchard 5 only. Samples were obtained from both commercial pickings, consisting of eight bushels in each type of wrap. These were equally divided, four of each being placed in store two days after harvest, the remainder after 14 days.

Results are given in Table 4.

TABLE 4. EFFECT OF PICKING DATE, DELAY IN STORAGE, AND TYPE OF WRAP. ROME BEAUTY, 1954

Orchard No.	Picking Date	Delay between Harvest and Storage (days)	Period between Harvest and Post-storage Examination (days)	Scald %	
				Plain Wraps	Oiled Wraps
5	20 April	2	111	0	0
	20 April	14	111	0	0
	7 May	2	101	2	1
	7 May	14	101	37	10

DISCUSSION

Although no scald developed on experimental fruit until after it was removed from store in either 1951 or 1952, it did appear on commercial stocks. It is of interest to note that all commercial stocks had been sold by the time the experimental samples were removed.

Although direct comparisons of fruit from the one orchard were not made, Frimley Beauty generally appears to be more susceptible than Rome Beauty to superficial scald (Table 1). Scald developed earlier in some years than in others. In the years 1951 and 1952, experimental fruit was not removed until the latter part of July, whereas in 1953 scald had developed in store by mid-June on Frimley Beauty and early July on Rome Beauty (Tables 1, 3).

Fruit of both varieties from certain orchards was more susceptible to scald than fruit from other orchards (Table 1). Scald development and susceptibility varied from year to year (Tables 1, 3). In general, fruit of the earliest picks (Table 2) scalded more than later picks. The amount of scald that developed in later picks varied, there being no consistent pattern of scald development.

Delay between harvesting and cool storage allowed more scald to develop in the variety Frimley Beauty than in Rome Beauty. When scald did develop it was moderately severe on fruit withheld from store for seven days and very severe if the period of delay was increased to 14 days (Table 3). Rome Beauty did not scald unduly if withheld from store for a week but periods of 14 days were injurious (Tables 3, 4).

Some samples of fruit remained free from scald (Tables 1, 3, 4). When scald did develop, wrapping the fruit in oiled paper reduced the incidence by 50% or more, whether the fruit was placed in store immediately after harvesting or was subjected to varying periods of delay. (Tables 1, 4). As it is not possible to predict whether a particular line of fruit will scald, the general adoption of oiled wraps for these varieties grown in Hawke's Bay would probably be fully justified on economic grounds. It is recommended that fruit of both these varieties should be wrapped in oiled paper.

COMPARISON OF FOUR ROOTSTOCKS FOR SULTAN PLUM IN THE AUCKLAND DISTRICT

By A. Farmer, Fruit Research Station, Department of Scientific and Industrial Research, Auckland

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Summary

An account is given of the vigour, size, and cropping, after seven years growth, of Sultan plum on Buck, Marianna, and Myrobalan B clonal plum stocks and on peach seedlings.

The largest trees were on Buck. Medium-sized trees were produced by Marianna and Myrobalan B. Peach gave smaller and more variably sized trees.

Total production of fruit for the first four years of cropping on Buck and Marianna was practically equal and was significantly greater than the production from trees on Myrobalan B and peach.

Under the conditions of the experiment, Marianna appears to be a more promising stock for the Japanese-type plums than the commonly used Myrobalan.

The value of Buck as a rootstock will depend on the relative production costs from very large compared with medium sized trees.

Introduction

The most important plums in New Zealand are the Japanese-type (*Prunus salicina*) or hybrids of this type with other species of plum. European-type plums (*P. domestica*) are not favoured by most commercial growers. General nursery practice is to bud plums on seedlings or cuttings of Myrobalan (Cherry plum). Marianna or Buck plum cuttings and peach seedlings are occasionally used as stocks.

Tree vigour, size, and cropping of Sultan (syn. Garnet, Occident) plum on four rootstocks planted at the Oratia Research Orchard are discussed in this paper. This trial, initiated by Mr. C. E. Woodhead (now Pip Fruit Specialist, Department of Agriculture) is continuing. While these trees can be expected to continue growing and to increase their production, the progress results presented here are those obtained after the first seven years' growth.

Material and Method

The experiment consisted of 40 trees laid out as 10 randomized blocks of 4 trees, including one tree on each of the following stocks (the first three were clonal):—

Buck

A sterile chance hybrid seedling imported into Australia from

England and sometimes used in New Zealand under the name Morrison's plum stock (Farmer 1953).

Marianna

Originating from Texas, U.S.A. and generally supposed to be an open-pollinated cross of *Prunus cerasifera* and *P. munsoniana.*

Myrobalan B

A seedling selection of *P. cerasifera* made by the East Malling Research Station, England (Hatton 1920).

Peach

Seedlings from pits of Golden Queen peach.

Sultan was selected for the scion variety as being typical of the red-fleshed group of Japanese-type plums which are important market varieties.

Trees were budded in 1947 and planted in 1948, 18 ft. apart on the square, in a solid block. The soil in the trial area is typically a mature podzol of low fertility, characterized by shallow clay loam on a heavy clay subsoil. Trees were pruned to an open centre similar to the method practised in the district. Variations in growth habit, caused by different rootstocks, necessitated slight modifications. Each tree was therefore treated according to its requirements. Clean cultivation was maintained during spring and summer and a complete commercial fertilizer was applied every year except 1952.

The normal annual rainfall for Oratia is 64 in.

Increase in tree growth was recorded annually by measuring the trunk girth six inches above the bud union. Trunk girth is considered to be the best indication of tree vigour. Height and spread were recorded only in the final year under review to give an overall picture of tree size. Figures given for spread represent the mean of readings taken in two directions at right angles.

Fruit crop was weighed in 1952, 53, 54, and 55.

RESULTS

The satisfactory crops produced with no provision for cross pollination indicate that Sultan is self-fertile.

Effect of Rootstock on Tree Vigour

Annual measurements of trunk girth of trees on the four rootstocks are shown in Fig. 1.

The average height, spread, and girth of trees on the four rootstocks, seven years after planting, is shown in Table 1.

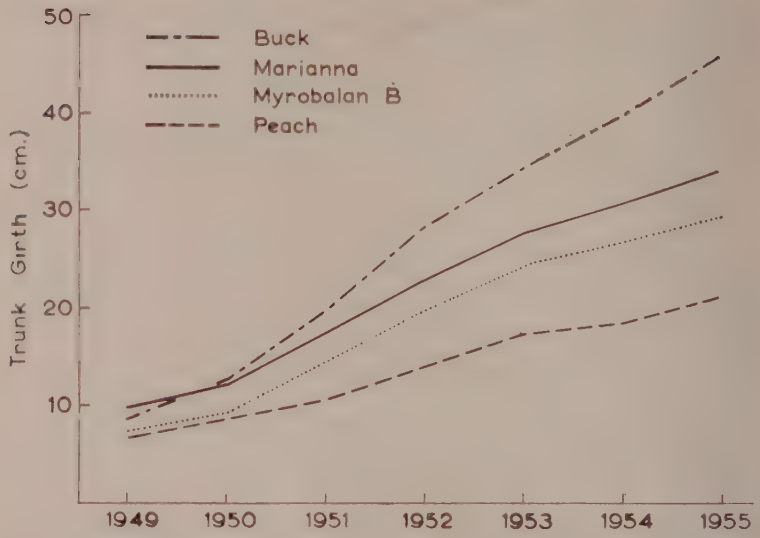

Fig. 1.—Annual girth measurements of Sultan plum on four rootstocks.

TABLE 1. AVERAGE HEIGHT, SPREAD, AND GIRTH OF SULTAN PLUM ON FOUR ROOTSTOCKS (PLANTED 7 YEARS)

Rootstock	Height (m.)	Spread (m.)	Girth (cm.)
Buck	3.3	5.1	45.9
Marianna	2.4	4.2	33.9
Myrobalan B	2.2	3.7	29.1
Peach	1.7	3.1	21.2
Sig diff. at 5%	0.3	0.5	4.3
at 1%	0.5	0.7	5.8

Considering trunk girth together with height and spread, trees on Buck are significantly larger than trees on the other stocks. Similarly, trees on Marianna are significantly larger than those on peach. Trees on Marianna show an increase in spread and girth (sig. at 5%) over trees on Myrobalan B. Trees on Myrobalan B are significantly larger than those on peach in height and girth (at 1%) and in spread (at 5%). Trees on the clonal plum rootstocks are fairly uniform, whereas trees on peach seedlings are variable. Some are pratically as large as trees on Marianna and Myrobalan B whereas others are very small and stunted.

Rootstock Effect on Cropping

Average annual crops per tree from 1952-1955 inclusive for each of the four rootstocks are shown in Fig. 2.

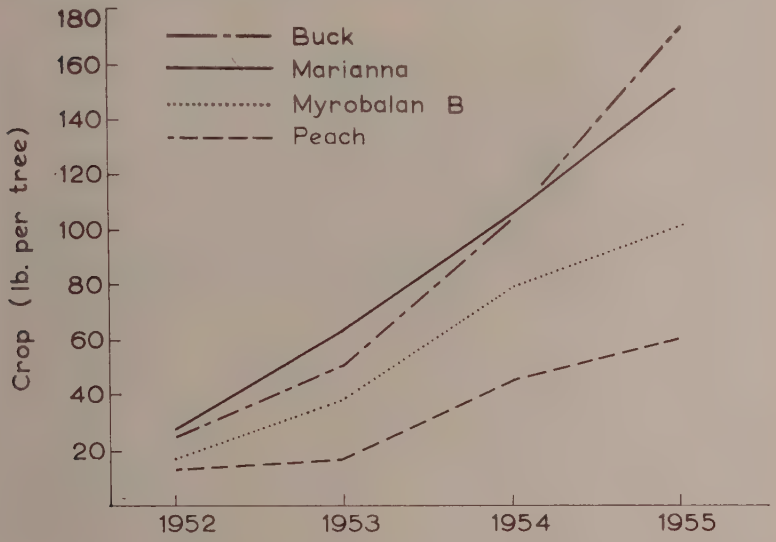

Fig. 2.—Annual crops of Sultan plum on four rootstocks.

The accumulated total crop per tree for each of the four rootstocks is shown in Table 2.

TABLE 2. TOTAL CROP OF SULTAN ON FOUR ROOTSTOCKS, 1952-1955

Rootstock	Crop (lb. per tree)	Rootstock	Crop (lb. per tree)
Buck	353.7	Myrobalan B	235.2
Marianna	346.0	Peach	132.0

Sig. diff. at 5%, 79.7; at 1%, 108.3.

Total production from trees on Buck and Marianna was significantly greater (at 1%) than from trees on Myrobalan B or peach. Total production from trees on Myrobalan B was significantly greater (at 5%) than from trees on peach.

CONCLUSIONS

The results indicate that with Sultan as scion variety, the rootstocks Buck and Marianna produce larger trees and heavier yields than Myrobalan B and seedling peach.

The performance of trees on Marianna is noteworthy. This stock produced medium-sized trees which commenced cropping early and have given high yields. Under these conditions Marianna appears to be a more promising stock for Japanese-type plums than the commonly used Myrobalan.

The vigour of trees on Buck is apparent, and because of the close correlation between tree size and cropping capacity it seems probable that these large, vigorous trees will continue to give outstandingly high yields. Before full use can be made of information from such rootstock trials, factors affecting the cost of production from small, medium, and large trees under New Zealand conditions must be investigated.

Acknowledgement

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PASTURE QUALITY AND RUMINANT DIGESTION

II. LEVELS OF VOLATILE ACIDS AND AMMONIA IN THE RUMEN OF SHEEP ON A HIGH-PRODUCTION PASTURE

By A. T. Johns, Grasslands Division, Department of Scientific and Industrial Research, Palmerston North

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Summary

The volatile acid and ammonia levels in the rumen of sheep grazing on high-production pasture have been determined throughout the year and compared with the concentrations found on a diet of hay. Maximum post-feeding ammonia-N levels varied between 35 and 130 mg. per 100 ml. of rumen contents. Differences were found in the percentage of individual acids, particularly when pasture was compared with hay. The possible relationship between these differences and animal metabolism are discussed.

Introduction

The efficiency with which domestic animals convert foodstuffs of varying chemical composition into meat, wool, and milk, has been studied for many years, but only during the last decade have serious attempts been made to link observed changes with the fermentation processes occurring in the rumen. The chief non-nitrogenous end products of ruminant digestion are acetic, propionic, and butyric acids, and the levels of these acids in relation to food supply on the one hand and animal performance on the other are being investigated by a number of workers. The demonstration by Popjàk *et al.* (1951) that acetate is a milk-fat precursor in the cow suggested that milk-fat content might be influenced by the rate of acetate production in the rumen. Previously Stoddard *et al.* (1949) had shown a marked decrease in fat content when cows received concentrates *ad. lib.* with less than 6 lb. hay daily, a decrease which could be counteracted by the administration of acetate. McClymont (1950) described various cases in Australia where grazing of low roughage pastures reduced the fat content of the milk. Balch (1952) found that a change in diet from long hay to ground hay was accompanied by a depression in the fat content of the milk and by a reduction in the total amount of acetate in the rumen. In a later paper Balch *et al.* (1952) pointed out that this depression in fat content of the milk frequently occurs when stall-fed cows are turned out to pasture in the spring. The inference from these investigations is that a decrease in, or change in character of, the fibre results in lowered acetate levels.

Of the other volatile acids present, propionic acid acts almost solely as a carbohydrate precursor (Kleiber *et al.* 1953) while butyric, qualitatively the largest source of energy within this fraction (Pfander and Phillipson 1953), is metabolized partly to carbohydrate and partly to fat (Kleiber *et al.* 1954). Butyric acid is known to be strongly ketogenic both in liver tissue and in the rumen epithelium (Pennington 1954). However, it seems likely that the fate of butyrate in the normal animal will be influenced by the level of propionate being simultaneously oxidized. Hence not only the level of butyrate but the butyrate—propionate ratio would be expected to be of importance in animal metabolism.

The C_5 acid fraction of the volatile fatty acids of the rumen has been shown by Annison (1954) to consist of n-valeric, isovaleric and 2-methyl butyric acids. He found some correlation between the branched-chain fatty acids and the quantity of protein fed. Interest in these branched-chain acids has increased through the recent finding by Shorland and co-workers (see Hansen *et al.* 1953) that they are constituents of ruminant fats, and the discovery by Bryant and Doetsch (1954) that some branched- as well as straight-chain fatty acids are necessary for the maximum growth of one of the important cellulolytic bacteria of the rumen.

The proteins of the feed are partly broken down by micro-organisms in the rumen and reassimilated as bacterial protein. The amount reassimilated by the microbial flora is dependent on the amount of readily fermentable carbohydrate present. McDonald (1948) first demonstrated that ammonia derived from the deamination of the amino acids of the food protein is an important component of the rumen ingesta, and that appreciable quantities may be absorbed into the blood draining the rumen. McDonald (1948) and Chalmers and Synge (1950) found that different proteins were attacked at different rates in the rumen; casein was rapidly deaminated whereas zein and herring meal were attacked more slowly. Herring meal was found by the latter authors to be a better protein supplement for sheep than casein, and the latter investigators were able to correlate this with the lower ammonia levels in the rumen and lower nitrogen excretion in the urine obtained with herring meal.

The rate of fermentation and total amounts of the individual volatile acids and ammonia produced cannot be accurately determined by direct sampling from the rumen, as absorption of these constituents through the rumen wall and dilution with saliva decreases their concentration simultaneously with their increase through fermentation. Moreover, the individual acids are not absorbed at the same rate. Pfander and Phillipson (1953) introduced into a sheep's rumen, which had been washed out and isolated, a solution of the sodium salts of the fatty acids in a bicarbonate saline medium. The ratio of acetic to propionic was approximately 3 : 1 but the proportions in which they were absorbed was approximately 2 : 1. Rates of production can be estimated by *in vitro* anaerobic fermentation of a substrate with rumen contents. However, unless elaborate precautions are taken, this fermentation dissociated from the absorption properties and secretory activities of the animal soon becomes abnormal. Despite these difficulties, both methods have been used by a number of investigators and have yielded very useful information.

The estimation of the level of the volatile acids and of ammonia in the rumen of sheep has almost invariably involved stall-fed animals. No long-term study has been made of grazing animals whose sole diet is pasture herbage. From the results presented in Part I and the evidence quoted above, even allowing for the limitations of the methods employed, it might be expected that volatile acid ratios would change with the seasonal changes in growth, and that ammonia levels would fluctuate with the seasonal change in protein content of the herbage.

In this paper are reported the total volatile acid levels, individual fatty acid percentages, and ammonia concentration, as determined by

sampling from the rumen of sheep grazing the pastures whose chemical composition was given in Part I.

METHODS

The pasture whose botanical and chemical composition was given in detail in Part I was grazed by sheep fitted with permanent rumen fistulae (Jarrett, 1948). The sheep were on a pasture of similar composition for at least 7 days before each grazing of the experimental area. The animals were placed in stalls without food overnight before the experimental period in order to obtain a sample of rumen contents before feeding, and at various times after the commencement of grazing.

Large samples (200 g.) of rumen contents were obtained through the fistula to minimize sampling error. The rumen liquid was strained through muslin to remove solid material.

Ammonia was determined by the method of Conway (1947) as applied to rumen contents by McDonald (1948). Total volatile acids were determined by adding to 5 ml. rumen contents, 1 ml. 10 N H_2SO_4 and 4 g. $MgSO_4.7H_2O$ followed by steam distillation in the Markham still. Quantitative recovery of the acids was obtained by collecting 50 ml. of the distillate. The distillate was titrated with 0.1 N NaOH under CO_2-free conditions.

Individual acids were determined by the gas–liquid chromatographic procedure of James and Martin (1952) run at 100°C. with the automatic titration unit. In certain analyses some doubt existed as to the exact ratio between n- and iso-butyric acids and in those cases total butyric acid only is quoted. Similarly the C_5 acids are grouped together as a single value.

RESULTS

The concentration of total volatile fatty acids (V.F.A.) and ammonia in the rumen contents of two sheep at approximately monthly intervals throughout the year are given in Table 1. The percentage contribution of individual fatty acids to total V.F.A. is also presented. Except where indicated, the first sample was taken before the animal was returned to pasture after being indoors overnight. At approximately two-hourly intervals during the day the sheep were removed from pasture for a few minutes to permit sampling. Although analyses were performed on all samples, values for the initial and one post-feeding sample only are given. The latter was chosen to fall within the period 5 to 8 hours after the feeding period commenced, when ammonia and V.F.A. concentrations were at or near maximum values. The course of fermentation is shown graphically in Fig. 1 for a winter and summer sample respectively.

In order to provide a basis of comparison with results published by other workers, experiments were also carried out with hay and hay-concentrate rations. In the first experiment, two sheep were changed abruptly from a good ryegrass–red clover hay (17.5% protein) to a ryegrass–white clover winter pasture (31% protein). After 4 days they were changed back to hay. The changes occurring in ammonia and V.F.A. levels are given in Table 2.

TABLE 1. THE VOLATILE FATTY ACID AND AMMONIA LEVELS IN THE RUMEN OF SHEEP AT PASTURE AT VARIOUS PERIODS OF THE YEAR

Date	Sampling Time	NH_3-N mg/100 ml		V.F.A. mM/100 ml		Acetic *		Propionic *		Isobutyric *		Butyric *		C5 acids *	
		I	II	I	II	I	II	I	II	I	II	I	II	I	II
17.8.53	0930†	65.1	59.0	4.9	6.3	54	59	27	20	5	5	9	13	5	4
	1500	71.6	80.6	7.5	14.3	54	50	28	30	3	3	9	13	7	4
15.9.53	0830	13.8	11.5	2.6	2.3	62	62	16	17			16**	13**	6	8
	1300	88.6	66.4	13.7	10.1	57	56	22	24			16**	15**	5	5
12.10.53	0845	45.2		6.0		61		15				18**		6	
	1600	99.1		13.5		55		23				17**		5	
12.11.53	0845	14.1	14.0	5.7	6.2	63	62	19	21	3	3	10	9	4	4
	1430	62.4	56.7	14.7	13.6	56	60	27	22	3	2	11	13	3	3
22.12.53	0930†	32.0	40.5	4.9	5.1	64	62	16	19	4	4	13	11	3	4
	1445	79.0	86.9	14.9	15.4	61	62	22	21	2	3	11	11	4	3
11.1.54	0900	31.9	40.1	5.0	4.5	59	57	19	20	4	4	12	12	6	7
	1445	91.3	86.4	15.5	15.3	58	60	23	23	3	2	12	13	4	3
11.2.54	0820	10.2	15.8	3.6	6.6	56	59	20	22	5	3	11	10	8	6
	1600	37.7	61.1	16.3	22.0	57	58	24	24	2	2	13	13	4	3
5.3.54	0820	22.0	20.4	4.8	4.4	63	60	13	16	5	6	12	12	7	6
	1430	44.3	35.2	14.4	13.0	56	59	24	23	3	3	10	9	6	6
2.4.54	0830	24.7	33.2	4.5	5.4	62	61	14	16	4	4	13	12	7	7
	1545	65.0	41.5	16.0	11.7	61	62	22	22	3	4	10	8	4	4
4.5.54	0830	19.7	20.0	3.2	3.0	59	55	15	17		4	17**	16	8	8
	1545	58.3	33.0	12.8	12.4	58	54	22	25	3	3	9	11	8	6
16.7.54	0845	36.4	33.6	3.9	4.3	53	54	20	19		5	20**	13	7	9
	1530	95.6	130.0	12.3	17.8	55	54	24	26	2	3	14	11	5	6
6.8.54	0830	20.7	31.0	3.9	6.5	57	50	16	21	5	5	12	14	10	10
	1330	100.0	94.5	16.5	18.7	58	54	21	22		2	16**	11	5	6

* Individual V.F.A. as a percentage of the total V.F.A.
** Combined butyric acids
† 1st sample 30 min. after start of feeding

TABLE 2. EFFECT OF TRANSFERRING SHEEP FROM HAY TO PASTURE ON THE VOLATILE ACID AND AMMONIA CONCENTRATIONS IN THEIR RUMEN

Date	Feed	Sampling Time	NH_3-N mg/100 ml		V.F.A. mM/100 ml		Acetic *		Propionic *		Isobutyric *		Butyric *		C5 acids *	
			I	II	I	II	I	II	I	II	I	II	I	II	I	II
5.7.54	Hay 17.5% protein	0830	23.8		5.8	6.9	69	65	19	17	3	3	7	10	3	5
		0930	29.7		9.7	11.5	66	65	21	24	3	2	6	6	4	4
		1045	25.0		9.7	11.0	68	67	20	18	4	3	6	7	2	5
		1315	22.3		9.9	10.5	65	66	22	20	2	2	7	7	4	5
		1515	30.1		13.8	12.6	65	67	24	21	3	2	5	7	3	3
6.7.54		0830	26.9	27.4	8.7	6.6	71	66	17	15	2	4	8	11	3	4
6.7.54	Put out to graze pasture 31% protein	0945	43.6	50.9	11.8	10.5	67	62	17	23	4	3	9	9	4	3
		1315	71.9	95.0	16.1	17.8	66	64	20	21			12**	10**	3	4
7.7.54	„	1515	94.0	139.0	15.6	21.6	54	51	25	25	3	3	15	17	4	4
9.7.54	„	0830	30.4	40.5	4.5	7.5	55	59	22	19	6	4	13	13	5	6
		1530	114.0	125.0	15.5	19.0	54	51	24	27	6	3	13	15	4	4
10.7.54	„	0830	45.7	77.9	9.4	11.3	55	57	21	22	4	3	14	14	6	5
10.7.54	Returned to hay	1530	41.3	40.1	14.5	12.1	63	61	26	25	2	2	7	12	2	2
11.7.54	„	0830			5.6	7.9	66	59	16	21	4	4	10	11	4	5
		1530			10.5	11.6	70	68	21	22	2	2	6	5	2	3
16.7.54	„	1600			10.7	12.7	72	72	19	19	2	2	6	7	1	1

* Individual V.F.A. as a percentage of the total V.F.A.

** Combined butyric acids

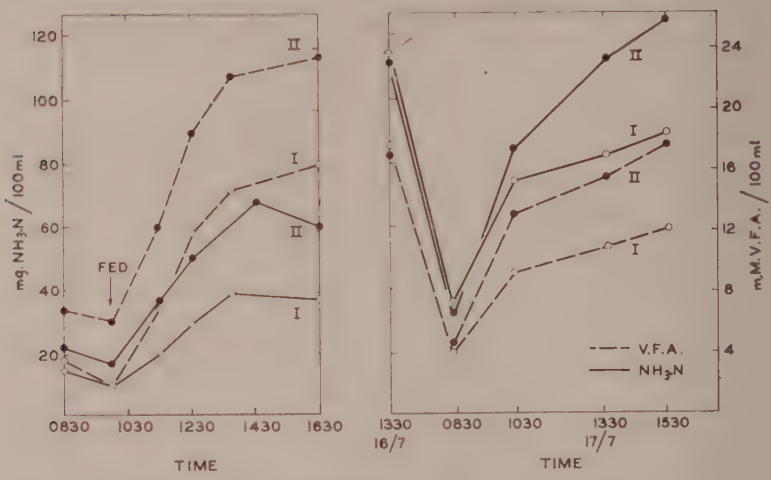

Fig. 1.—The increase in volatile fatty acids and ammonia in two sheep. *Left*: On a summer pasture. *Right*: On a winter pasture.

In the second experiment, hay of 12.9% protein content was compared with a ration of hay, crushed oats, and concentrate (17.3% protein). Detailed results are not presented as they merely confirm those in Table 2 and in the reports of other workers.

Formic acid has been reported (Gray *et al.* 1952) to be present in concentrations as high as 5% of the V.F.A. fraction. In over four hundred estimations of the individual acids in the rumen contents of sheep on various diets, formic acid was never detected. On several occasions a known amount of formic acid was added to the sample and quantitative recoveries obtained.

Discussion

The results presented above represent a preliminary survey, using only two sheep, to test the practicability of the method. Differences between the animals in respect to all analyses are relatively large (see Fig. 1) and it is obvious that only by using larger numbers can the significance of concentration differences be assessed. As an illustration, the results obtained from Sheep I support the finding of Gray and Pilgrim (1951) and of Shazly (1952), viz. that the contribution of acetic acid to V.F.A. is higher, and of propionic acid lower, before feeding than after. This conclusion could not be derived from the behaviour of Sheep II.

Table 1 shows no marked change in total V.F.A. or in acid proportions with changing pasture composition throughout the year. The maximum range in V.F.A. was from 10.1 to 18.7 mM/100 ml. and, in percentage contributions, acetic acid 50–62%, propionic 21–30%, butyric acid (total) 12–17%, and C_5 acids 3–10%. Phillipson (1952) quotes, for grass fed sheep, acetic acid 62–63%, propionic 17–19%, butyric 13–15%, and higher acids 5–7%. Shazly (1952) fed frozen and dried grass prepared from the same pasture herbage (22% protein).

For frozen grass, acetic acid values ranged from 54–55% of V.F.A., propionic from 23–24%, and butyric from 14–17%; for dried grass, the corresponding values were 58–64%, 22–26%, and 9–12%.

The largest variations in proportion occurred when the animals were changed from hay to pasture herbage and then back to hay. Acetic acid proportion dropped markedly on pasture and this was accompanied by an increase in total butyric acid. Propionic acid percentages were relatively constant. The level of volatile acid at the peak of fermentation was in general higher on pasture than on hay, which contrasts with the findings of Carroll and Hungate (1954). High acetate percentages were also obtained on a hay of poorer quality and on a ration of hay and crushed oats.

The only other results (of which the author is aware) of a study of changes in the ratio of V.F.A. in the rumen with seasonal changes in pasture are those of Card and Shultz (1953). In this case, however, hay and silage were made from the same field at different cutting dates. They found significant differences between cows fed equal amounts of grain and legume–grass hay or silage, according to its date of cutting. Roughages from the later dates of cutting tended to cause an increase in acetic and a decrease in butyric. Silage tended to cause a decrease in acetic and an increase in propionic when compared with hay. The general tendency in all their studies was for acetic to be the most variable, with reciprocal changes taking place in butyric.

Our results were somewhat similar and differ from the changes found by Balch (1952) on changing from long hay to ground hay where decrease in fat content of the milk was accompanied by a marked lowering of the percentage of acetate in the rumen fluid and a reciprocal increase in propionate.

In our present state of knowledge it is impossible to evaluate the significance of these fatty-acid differences in terms of animal nutrition. In the introduction, reference was made to a number of studies in which changes in the V.F.A. fraction were related to various aspects of animal metabolism, but many quantitative investigations are required before satisfactory generalizations are possible. Similarly, the relationship between feed and end products of ruminant digestion awaits much more accurate definition of feed composition together with a more precise knowledge of the fermentation reactions to which its individual components are subjected. In the present study, considerable variation occurred in certain chemically defined constituents of the herbage ingested by the animals throughout the year, but these appear to bear no relation to the observed changes in V.F.A. levels and proportions. This however may be no more than a reflection of the inadequacy of the technique employed, and the use of *in vitro* studies is desirable before definite conclusions can be drawn.

The high protein levels in the pasture herbage are reflected in high ammonia levels in rumen contents. The range of concentration was from 35 to 130 mg./100 ml., but peak levels did not coincide with peak protein levels in the herbage, nor did low concentrations necessarily result from the ingestion of low-protein herbage. It was shown in Part I that the N concentration of ryegrass herbage increased markedly with increasing height above ground level. Coupled with the selective grazing habit of the sheep this variation causes difficulty in estimating the

protein content of the herbage being eaten. Nevertheless, in spite of these limitations, it might have been expected that rumen ammonia levels would follow pasture nitrogen levels more closely. Recent investigations by the Rowett group (c.f. Shazly, 1952, and Chalmers and Synge, 1954) show that the fermentation of protein by ruminants depends on the protein used, the degree to which its digestibility has been changed by pre-treatment, and the type and level of carbohydrate being simultaneously fermented. A much more precise definition of both nitrogenous and non-nitrogenous constituents of herbage is obviously necessary before the prediction of rumen ammonia levels can be made with confidence.

The high ammonia levels recorded in winter and early spring are of interest in relation to mineral metabolism in the grazing animal. Herbage during the period is characterized by Melville and Sears (1953) as being high in protein, high in potassium (up to 4% of the D. M.), and low in calcium. As a large proportion of the potassium and ammonia is absorbed into the blood stream, the influx of these ions may depress the intake of other cations. A number of experiments have been reported in which changes in levels of potassium intake produced no effect on the blood levels of magnesium and calcium (Pearson *et al.* 1949). On the other hand Kunkel *et al.* (1953) showed that the inclusion of 5% of potassium as bicarbonate in the diet resulted in a significant lowering of serum magnesium. The calcium, potassium, and sodium levels were not markedly altered.

Phillipson (1953) states that rumen liquor contains 99 to 175 mg. % of potassium. A concentration of 100 mg./100 ml. ammonia-N represents 121 mg. ammonia per 100 ml., while the highest value recorded in Table 1, viz. 130 mg./100 ml., corresponds to 158 mg. ammonia. Hence the rumen levels of ammonia on high-production pasture herbage rival those of potassium and it would be expected that any adverse effect caused by the latter would be much enhanced.

Other deleterious effects may also result from high rumen ammonia levels. Thus Edson (1953) found that the production of acetoacetic acid by liver slices is increased in the presence of ammonium chloride, while Huennekens *et al.* (1951) reported on the inhibitory effect of ammonia upon propionate metabolism by the liver "cyclophorase" system.

Pennington (1954) observed that ammonium chloride partially inhibited the uptake of propionate by rumen epithelium, while no such effect could be observed with butyrate. The magnitude of the inhibition of propionate metabolism varied consideraly from experiment to experiment, but very considerable effect was experienced at 0.03 M NH_4Cl. In the present work, levels of ammonia greatly in excess of this concentration were encountered (105 mg. ammonia N/100 ml. is 0.075M). The inhibition of propionate metabolism could have the effect of removing its antiketogenic effect on the associated butyrate conversion to ketone bodies. With the high-protein diet of winter grass it is likely that better utilization and better stock thrift would result from the supplementary feeding of a readily fermentable carbohydrate.

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THE UPTAKE OF PHOSPHORUS BY THE SHEEP

I. THE EXCRETION OF INJECTED RADIOPHOSPHORUS
II. THE AVAILABILITY OF THE PHOSPHORUS OF A DIET OF HAY AND CONCENTRATES

By EVAN WRIGHT, Animal Research Station, Department of Agriculture, Wallaceville

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Summary

1. An investigation of the excretion of injected radio-phosphorus has been made, and it has been shown (a) that there is a variable time lag between similar events in the faecal and urinary excretion, (b) that there is a daily rhythm of excretion, and (c) that the specific activity of the urine may be used as an estimate of the endogenous faecal specific activity.

2. By the use of the urinary specific activity to calculate endogenous faecal phosphorus, the uptake of dietary phosphorus has been found to vary widely from day to day, and from sheep to sheep, with a maximum of 100%. The only significant correlations between intakes and excretions are between absorption of phosphorus and phosphorus balance. This is associated with a regression co-efficient of approximately one. It is postulated that, under the conditions of the experiment, the state of phosphate balance is determined not by excretion but by control of absorption by unspecified factors.

INTRODUCTION

To determine whether sheep require a phosphorus supplement, it is important to know the extent to which the phosphorus of the diet is available to the animal. (Cunningham 1947; "Researcher" 1947).

A cycle of absorption and re-excretion makes the measurement of phosphorus uptake difficult, as the phosphorus appearing in the faeces is partly unabsorbed dietary phosphorus, and partly phosphorus that has been absorbed and re-excreted into the gut. Since Hevesy *et al.* (1939) introduced labelling with radioactive phosphorus to measure endogenous faecal phosphorus, several other workers have used this method (Kjerulf-Jensen, 1941; Kleiber *et al.*, 1951; and Lofgreen and Kleiber, 1953, 1954) while Wallace *et al.* (1951) and Comar *et al.* (1953) have employed a similar method for measuring endogenous calcium.

It is assumed that the endogenous material to be measured comes from a "pool", which can be labelled with radioactive isotopes, and whose consequent specific activity can be measured. For this to be true the pool must be in a state of equilibrium with respect to the active and stable isotopes of the element under investigation.

If the plasma is taken as the pool, a few hours delay after administration of radio-phosphorus suffices for equilibrium to be established (Govaerts 1947). If the urine is used to estimate the endogenous specific activity, it is necessary to take into consideration pools other than the plasma, since it is possible that, in the sheep, urinary phosphorus originates in the tissue of the urinary tract rather than in kidney excretion (Watson 1933). The turnover of phosphorus can probably be divided into two parts, one consisting of systems which reach equilibrium very rapidly, and the other of systems with very slow rates.

Such systems are plasma–extracellular fluid, and intracellular fluid—extracellular fluid, which have been shown by Ennor and Rosenberg (1954a) to be fast and slow respectively. The fast mechanisms come to equilibrium in, at most, a few hours. For example, in dogs, Govaerts (1947) found equilibrium between blood and urine in $1\frac{1}{2}$ hours and Weissberger and Nasset (1942) between blood and liver in $2\frac{1}{2}$ to $3\frac{1}{2}$ hours. In rats, while the exchange between blood and nervous tissue is very slow, equilibrium within the brain after injection of ^{32}P took only 8 hours (Dawson 1951; Lindberg and Ernster 1950). Stoner (1950) showed that plasma specific activity was constant in rats after 3 hours, while Gilbert, Janney and Hines (1950) assumed equilibrium between plasma and skeletal muscle in 16 minutes. The only results available for large animals, apart from dogs, are those of Kleiber, Smith and Ralston (1950) who showed that, in cows, the fast part of the removal of ^{32}P from plasma, which they suggest was to extra-cellular fluid, had a rate of 100% in 20 minutes.

The hypothesis that all pools, whose rates of turnover are fast enough to influence the relationship between endogenous faecal and urinary specific activities, are in equilibrium after three or four days, is supported by the work of Cohn and Greenberg (1938). They showed that, in rats, equilibrium between bone, carcass, muscle, blood, liver, stomach, and intestine was established in about 12 hours. Ennor and Rosenberg (1954b) found equilibrium between the muscle and plasma of rabbits in 90 hours, and Feaster, Shirley, McCall and Davis (1953) found that, in rats, equilibrium was reached between kidney, femur, liver, and blood by four days, and possibly earlier as no observations were recorded for the second and third days.

This paper describes experiments designed to study the availability of dietary phosphorus to the sheep by labelling the endogenous faecal phosphorus with ^{32}P. The results obtained from this investigation are discussed in Section 2. Section 1 contains some observations on the excretion of radiophosphorus, obtained in the course of the experiments mentioned above, with special reference to the usefulness of the urinary specific activity in the estimation of endogenous faecal phosphorus.

I. The Excretion of Injected Radio-phosphorus

Experimental Methods

Sheep

With the exception of one crossbred Romney wether, the sheep used were flock Romney ewes. They were housed in single pens and fed 1 kg. chaffed ryegrass and clover hay and 250 g. sheepnuts daily, given in one feeding at 6 p.m. Twelve-hour samples (except sheep 1 which was sampled at more irregular intervals) were obtained by the methods of Reid and Wright (unpublished), which ensure that the urine is not contaminated by faeces. In these methods urine is obtained from the ewe by permanent catheterization, and from the partially restricted wether through a funnel. In both cases the urine collects in a polyethylene tank slung below the belly and the faeces in a polyethylene-lined canvas bag.

Radioactive Phosphorus

The A.E.R.E. Harwell supplied the ^{32}P as orthophosphate, either carrier-free or made up, with added carrier to the extent of 100 μg. P. per ml., in isotonic saline. In the former case, carrier, to the extent of 100 μg/ml., was added at this Station. The very low ovine urinary phosphorus governed the radio-phosphorus dose rate, which was increased from 1 mc. for Sheep 1 to 2 mc. for Sheep 2, and 10 mc. for Sheep 3 and 4.

The radioactive phosphate was administered at 6 p.m. via the jugular vein, by injection into the delivery tube of a saline drip.

Preparation of Samples

The collected faeces were well mixed and about 150 g. were dried in an oven at 105°c. for 24 hours. A delay of 12 hours before weighing and bottling allowed the sample to come to equilibrium with air so that moisture content did not change appreciably during storage. A macerator with a sealable stainless steel container reduced the dried pellets to powder. It was necessary to carry out this operation in a gloved box, as the fine powder, which spread rapidly on opening the container, could cause dangerous contamination of the laboratory. Urine, removed from the collection tank to a graduated separating funnel by suction, was acidified by adding approximately 10% of its volume of concentrated nitric acid, a few drops of amyl alcohol preventing frothing. It had been found that, whether or not the acid was added, a precipitate formed on storage. In the former case the precipitate dispersed readily in the supernatant liquid for sampling, but in the latter it did not, and its flocculent nature prevented reproducible sampling.

Analysis

Methods of analysis used were:

(1) A 1 g. sample of faeces or a 50 ml. aliquot of urine (the large volume being necessary because of the low phosphorus level in ruminant urine) was wet digested, the phosphorus precipitated as molybdate and then as magnesium ammonium phosphate (MacKenzie and Dean 1948), the total P being calculated from the former and the radioactivity counted in the latter. This method was used for all urines and for faeces of Sheep 1.

(2) Faecal ^{32}P was measured by the infinite layer method of MacKenzie and Dean (1950). In this method, dry plant material is pressed into a briquette of thickness greater than the critical thickness for complete absorption of the radiation from the bottom layers. In the case of faeces it was necessary to grind the material, since it was not possible, using whole pellets, to produce a briquette with a flat surface. A sample of 4.5 to 5g. was measured by volume, the weight for critical thickness having been determined as approximately 2g.

The method of MacKenzie and Dean (1948) described above for urine analysis was taken as the standard method, and all estimations by method (2) were corrected by means of a conversion factor, obtained by carrying out both analyses on the same sample. The results are shown in Table 1, the mean being the value used in calculation for all experiments.

TABLE 1. CONVERSION FACTORS

Experiment	Sample	No. of Replicates	Conversion Factor
2	1	4	0.654
3	1	2	0.617
	2	2	0.600
	3	2	0.608
	4	2	0.658
4	1	4	0.643
	2	4	0.696

Weighted mean 0.654 ± 0.006

(3) Total faecal phosphorus was estimated by the Briggs modification of the Bell-Doisy procedure, after rapid digestion by the method of Bolin and Stamberg (1944). Methods (2) and (3) were used for all faeces other than those from Sheep 1.

Radioactivity was measured by a General Electric GM4 end-window counter mounted in a $1\frac{1}{2}$-in. lead castle. Most samples were counted to a standard deviation of 0.8%, the exceptions being the later urines from Sheep 1 and 2, which were counted to a standard deviation of 1.5%.

All counts were corrected for background, dead-time, and counter drift.

RESULTS

Excretion figures for four sheep are given in Table 2. Column one shows the period of collection taken from the time of injection of the radio-phosphorus for Sheep 1, while the second and third columns give the rate of excretion in urine and faeces, in counts per second excreted per hour. Similarly column 4 gives the time for other sheep and is followed by the urinary excretion and specific activity and then by the faecal excretion of each sheep.

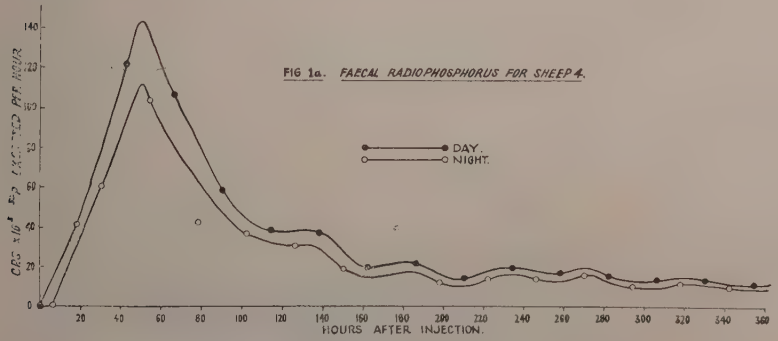


Fig. 1a.—Faecal radiophosphorus for Sheep 4.

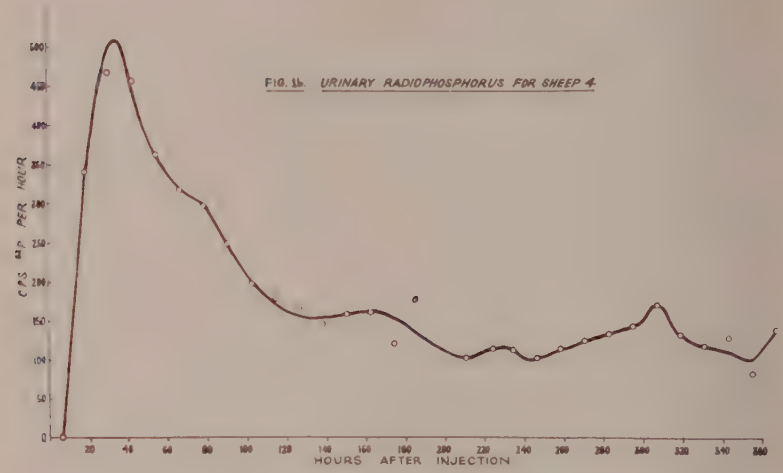


Fig. 1b.—Urinary radiophosphorus for Sheep 4.

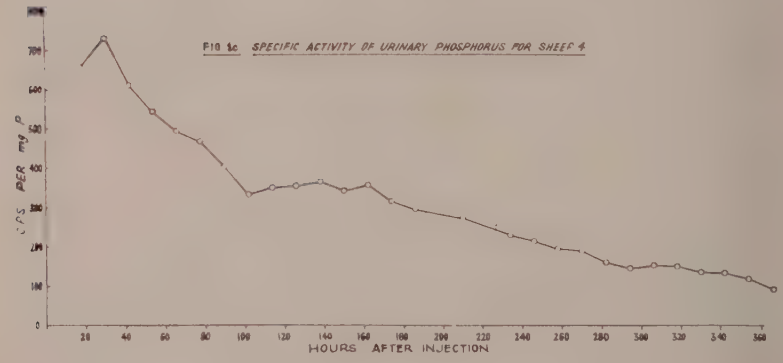


Fig. 1c.—Specific activity of urinary phosphorus for Sheep 4.

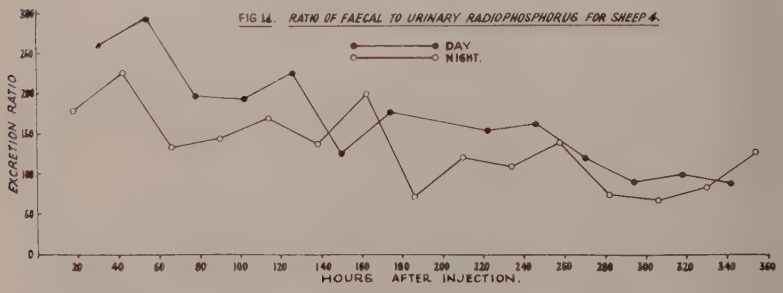


Fig. 1d.—Ratio of faecal to urinary radiophosphorus for Sheep 4.

TABLE 2.

Time in Hours	Sheep 1		Time in Hours	Sheep 2			Sheep 3			Sheep 4		
	Urine	Faeces		Urine		Faeces	Urine		Faeces	Urine		Faeces
	^{32}P cps/hr	^{32}P cps/hr		^{32}P cps/hr	Sp.Act. cps/mg	^{32}P cps/hr	^{32}P cps/hr	Sp.Act. cps/mg	^{32}P cps/hr	^{32}P cps/hr	Sp.Act. cps/mg	^{32}P cps/hr
0 – 48	13.3	609	0 – 12	11.65	17.6	271	24,600	2,097	27,300	—	—	216.2
48 – 72	14.3	7,754	12 – 24	36.19	46.7	36,080	3,460	553	158,400	341	659	41,270
72 – 89	17.2	2,823	24 – 36	38.29	54.2	20,110	12,840	1,010	174,700	468	732	60,660
89 – 97	18.9	7,131	36 – 48	29.15	49.3	22,150	3,770	972	212,100	457	613	122,200
97 – 113	14.9	3,856	48 – 60	17.64	33.2	12,300	8,010	757	137,000	364	546	103,500
113 – 121	20.4	2,886	60 – 72	11.73	15.7	10,990	7,640	992	125,100	318	496	107,000
121 – 137	15.8	2,434	72 – 84	15.17	25.5	6,520	5,910	441	115,200	298	470	42,680
137 – 145	17.4	1,941	84 – 96	15.60	23.9	6,378	6,350	486	81,310	250	407	59,110
145 – 161	11.8	1,933	96 – 108	12.92	23.5	3,104	—		56,770	199	332	36,700
161 – 169	11.1	2,246	108 – 120	16.56	21.2	5,582	5,500	474	41,660	180	350	38,550
169 – 185	12.4	1,705	120 – 132	9.98	17.4	3,070	7,790	421	53,680	166	356	30,410
185 – 193	15.8	2,220	132 – 144	9.72	12.4	2,640	2,070	259	36,770	147	368	37,510
193 – 209	—	1,467	144 – 156	11.51	17.4	1,700	2,890	222	35,720	159	342	18,920
209 – 211	9.4	1,809	156 – 168	9.85	13.0	3,390	2,200	205	28,220	162	359	19,960
211 – 233	12.8	1,133	168 – 180	6.57	11.3	1,570	2,500	192	27,290	122	317	32,370
233 – 242	13.8	2,331	180 – 192	9.26	14.1	2,700	2,290	189	17,740	168	298	21,470
242 – 257	7.6	1,581	192 – 204	5.56	13.5	1,520	3,040	221	11,920	—	—	11,980
257 – 265	16.6	1,900	204 – 216	7.47	12.4	2,490	2,800	198	15,170	104	276	14,300
265 – 281	6.2	926	216 – 228	5.36	11.9	1,490	1,870	150	21,540	126	246	13,480
281 – 290	14.3	969	228 – 240	7.69	12.8	2,160	2,260	157	21,830	124	230	19,360
			240 – 252			1,320	3,280	132	11,170	104	214	13,510
			252 – 264			1,670				116	196	16,810
			264 – 276			1,110				126	190	15,880
			276 – 288			1,550				135	161	15,420
			288 – 300			899				145	148	10,000
			300 – 312			766				172	153	13,510
			312 – 324			776				134	151	11,450
			324 – 336			1,190				120	138	13,550
			336 – 348			614				120	134	9,908
			348 – 360			1,020				84	120	11,420
			360 – 372			629				940	95	10,620
			372 – 384			954						
			384 – 396			777						
			396 – 408			776						

In Fig. 1 is a set of curves illustrating the phosphorus excretion of Sheep 4. Figs. 1a and 1b are plots of the faecal and urinary figures, respectively, of Table 2. Fig. 1c is a curve of urinary specific activity against time, while in Fig. 1d is plotted the ratio faecal ^{32}P/urinary ^{32}P allowing for the time lag between faecal and urinary excretion as described below. For the purpose of determining this lag, smooth curves have been fitted by eye to a and b and it is not intended that they should be used for other interpolations.

DISCUSSION

Daily Rhythm

In some cases plotting the excretion rate against time revealed an alternating pattern due to difference between day and night values.

The phenomenon of daily rhythms in metabolism has been well recognized and, in particular, has been demonstrated in phosphate metabolism of the liver by Ekman and Holmgren (1947), and in ruminant digestion by Kane, Jacobson, and Moore (1952) who observed a diurnal variation in the excretion of chromium and lignin by the cow. Both urine and faeces showed this rhythm in the present experiments. The faecal output of radio-phosphorus showed the variation in all cases, while two sheep showed it in the urine as well. In Sheep 3, which was one of those showing the rhythm in the urine, the nocturnal excretion rate exceeded the diurnal for a period of approximately 3 days during the experiment. This was in contrast to all other cases where the diurnal rate was the greater. In view of the work of Howard (1953), who found that these rhythms are closely related to diet, this inversion probably has no special significance. In the faeces, the regular pattern of Fig 1a was found in Sheep 2 and 4, and from 150 hours onward in Sheep 1. In Sheep 2 and 4 the mean ratio of diurnal to nocturnal excretion was calculated by comparing the day (or night) value with the mean of the preceding or following night or day values. That is to say, if N(n) is the n^{th} observation of the night rate and D(n) the n^{th} day-time value, the ratio is:—

$$R(n) = \frac{D(n-1) + D(n+1)}{2N(n)}$$

$$\text{or } R(n+1) = \frac{2D(n+1)}{N(n) + N(n+2)}$$

The mean value of R for Sheep 2 was 1.51 ± 0.05, and for Sheep 4, 1.26 ± 0.06.

Urinary Excretion

Martin and Pierce (1934) reported urinary phosphorus in sheep varying from "negligible" to 18 mg./100 ml. and Watson (1933) gives the figure 1 mg./100 ml. In the present experiment, Sheep 2 and 4 gave urinary phosphorus of approximately 1 mg./100 ml. while sheep 3 varied over the range 10–40 mg./100 ml. According to Watson there is no true excretion of phosphorus in the urine of the sheep until the serum phosphorus reaches 7.5 mg./100 ml., and such phosphorus as is found in the urine is due to the break-up of epithelial cells of the urinary tract. On this basis it is possible that the comparatively large value shown for Sheep 3 may have been due to tissue injury by the catheter.

Fig. 1c shows the curve for the specific activity of the urinary phosphorus for Sheep 4, Fig. 2a and 2b for Sheep 2 and 3.

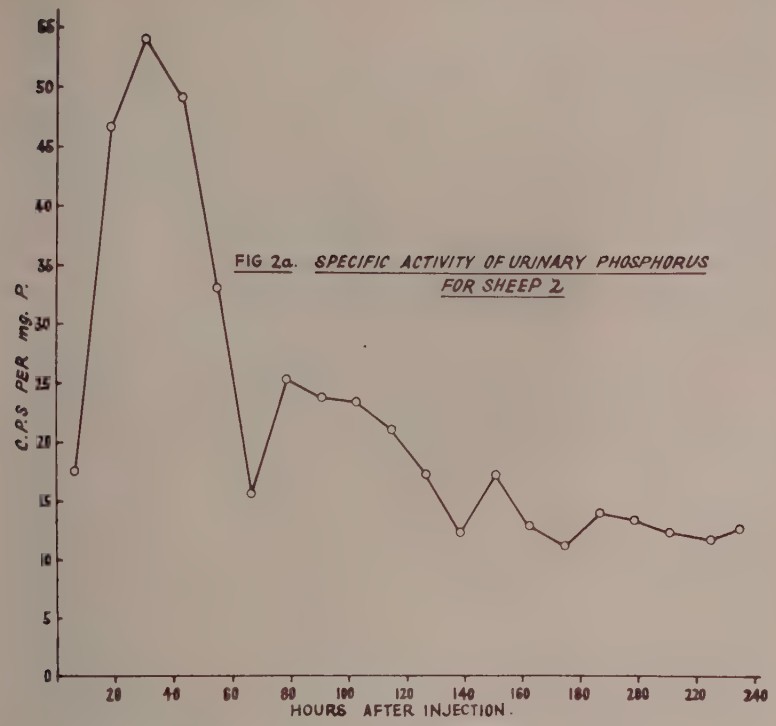
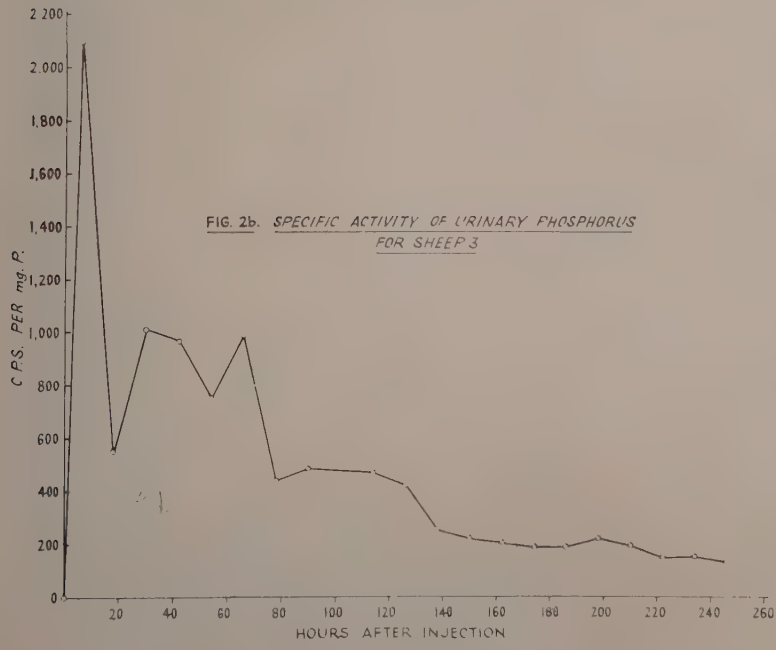


Fig. 2a.—Specific activity of urinary phosphorus for Sheep 2.



Fib 2b.—Specific activity of urinary phosphorus for Sheep 3.

An exponential curve of the form

$$\log\alpha = a + bt$$

where α is relative specific activity (with peak specific activity = 100); a is a constant; b the regression coefficient; and t time, fits the figures well. Values for b are given in Table 3, together with correlation coefficients. It is also of interest to know whether the pattern of urinary specific activity varies from sheep to sheep. The analysis in Table 3 shows that there is very little chance that these three regressions are estimates of the same parameter.

TABLE 3. REGRESSION OF LOG URINARY SPECIFIC ACTIVITY ON TIME

	Regression Coefficient (b)	Correlation Coefficient	Degrees of Freedom
Sheep 2	—0.0028 ± 0.0004	—0.85	17
Sheep 3	—0.0043 ± 0.0004	—0.95	18
Sheep 4	—0.0022 ± 0.0001	—0.99	27

ANALYSIS OF ERRORS OF ESTIMATE

Source of Variation	Degrees of Freedom	Sum of Squares	Mean Squares
Deviation from mean regression	61	0.7792	
Deviation from individual sheep regressions	59	0.4389	0.007439
Differences among regressions	2	0.3403	0.1702

F = 22.9 (F for significance at 1% probability level 4.9)

Temporal Relationship of Urinary and Faecal Excretion

In any method of relating intestinally secreted to faecal material, it is necessary to allow for the time taken for the secreted material to pass through the gut and be voided in the faeces. The notation Δt is used for this time lag in the following discussion. Kleiber and co-workers (Kleiber *et al.* 1951: Lofgreen and Kleiber 1953, 1954) have taken a standard time of 2 days for a cow and 1 day for a sheep. This lag was between the peak of activity in the blood and that in the faeces, whereas in the present work the lag was between the peaks in the urine and in the faeces. However, it is to be expected that any variation in this time lag would occur in the passage of the endogenous phosphorus through the intestine, rather than in the urinary excretion. In the present work the time lag between urinary and faecal excretion was extremely variable from sheep to sheep.

An inspection of the curves of Fig. 1a and 1b shows that the initial peak of activity is not the only maximum occurring. There are in fact a series of maxima in both urine and faeces. These are listed in Table 5. The second maximum does not fit the pattern of the others and has not been included in calculating the mean lag. None of the figures for Δt, except perhaps the first, are sufficiently accurate to stand alone,

but collectviely confirm the estimate of Δt made from the first maximum. The mean values of Δt, taken to the nearest multiple of twelve, and used for each sheep in further calculations, are given in the last line of the table.

Quantitative Relationship of Urinary and Faecal Excretion

The ratio of faecal to urinary radio-phosphorus is plotted against time in Figs. 1d, 3a, and 3b. This ratio is calculated from the faecal

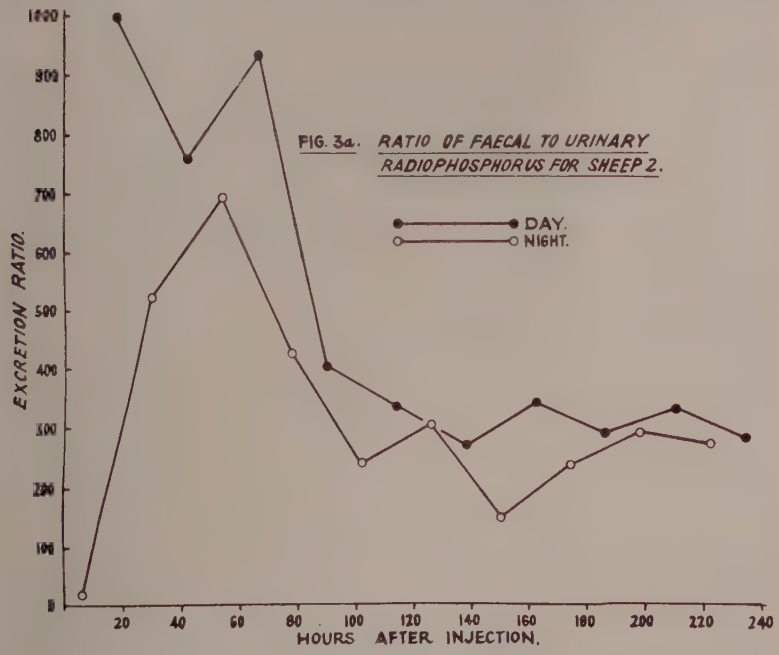


Fig. 3a.—Ratio of faecal to urinary radiophosphorus for Sheep 2.

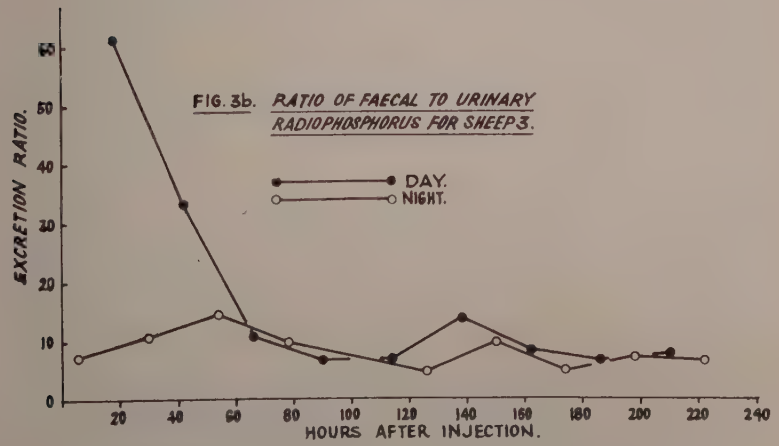


Fig. 3b.—Ratio of faecal to urinary radiophosphorus for Sheep 3.

radio-phosphorus at time t + Δt and the urinary radio-phosphorus at time t. These curves show a daily rhythm as well as a tendency to decrease with time. The differences between the three gradients are not significant at the 5% probability level and it might be assumed that they are expressions of a fundamental faecal-urinary relationship. However, a comparison with the corresponding gradients for total urinary phosphorus (Table 4) shows that while in Sheep 2 the two quantities are independent they are closely related in Sheep 3 and 4. Though the correlations are not significant in Sheep 3 they might be so were the varience due to daily rhythm removed.

TABLE 4. TIME REGRESSION OF THE RATIO OF FAECAL AND URINARY ^{32}P AND OF TOTAL URINARY P.

Regressions are given as percentage of mean per hour

	Faecal ^{32}P/Urinary ^{32}P		Total Urinary P.	
	Regression Coefficient	Correlation Coefficient	Regression Coefficient	Correlation Coefficient
Sheep 2	—0.16	—0.35	$<10^{-3}$	0.02
Sheep 3	—0.09	—0.13	0.03	0.27
Sheep 4	—0.30	—0.73**	0.37	0.734**

** Significant at 1%

TABLE 5. TIME RELATIONS OF MAXIMA IN FAECAL AND URINARY EXCRETION CURVES

Time after Injection	Sheep 2			Sheep 3			Sheep 4		
	Urine (1)	Faeces (2)	Δt (2)-(1)	Urine (1)	Faeces (2)	Δt (2)-(1)	Urine (1)	Faeces (2)	Δt (2)-(1)
Peak	27	20	—7	10	35	25	35	50	15
2nd Max.	80	120	—	?	78	—	80	135	—
3rd Max.	152	160	8	120†	126†	6	162	180	18
4th Max.				157	174	17	227	235	8
5th Max.				205	230	25	265?	270	5
6th Max.							305	320	15
Mean Δt			0			24			12

† by comparison with Sheep 2 and 4, the relationship of these maxima is doubtful. Including this figure, the mean Δt for sheep 3 is 18; excluding, it 22. In view of this, and since it is necessary on account of the 12 hourly sampling to take Δt to the nearest 12 hours, the figure 24 hours has been used for Sheep 3.

The Use of Urinary Specific Activity in the Estimation of Endogenous Faecal Phosphorus

The pool specific activity may be measured in either the plasma or the urine. The use of the urine has the advantage, not only of ease of collection, but also of providing a sample collected over the same

period as the faeces, and integrated with respect to time. On the other hand, the plasma can only give instantaneous values and it is necessary to give repeated injections of radio phosphorus in order to keep the blood level approximately constant.

Against the use of the urine is the possibility that its specific activity does not reflect that of the plasma. But, as discussed in the introduction, this would not be expected from purely theoretical considerations and, indeed, Lofgreen and Kleiber (1954) opposed its use, not because it showed a trend different from that of the plasma, but because it was too irregular. This, however, is more likely to be due to experimental error than to any fundamental cause, and it is not supported by the figures reported above.

It is unfortunate that, as the experiments were carried out before the publication of Lofgreen and Kleiber, no direct test was made, but, since the specific activity curve was reasonably regular, and its use to determine endogenous excretion leads to self-consistent results as described in Section 2, it is unlikely that such a test would have given an adverse result.

II. The Availability of the Phosphorus of a Diet of Hay and Concentrates

To find the percentage of dietary phosphorus absorbed, it is necessary to calculate the endogenous faecal phosphorus from the radioactivity of the faeces and the estimated specific activity of the re-excreted phosphorus. The procedure is summarized by the equation:—

$$A(t) = D(t) - \left(F\ (t + \Delta t) - \frac{f\ (t + \Delta t)}{\alpha\ (t)} \right)$$

There A is amount of dietary phosphorus absorbed, D is dietary intake of phosphorus; F is total faecal phosphorus; f is faecal radio-phosphorus; α is specific activity of urine; t is the mean time of collection of urine; and Δt is the time lag of voiding of faeces.

It is assumed that there is no time lag between feeding and urine excretion. Though this assumption cannot be strictly justified, the rapid absorption of phosphate (Wright, 1955) and equally rapid equilibration of exogenous blood phosphate (Govaerts 1947), together with the probability that the endogenous faecal phosphorus originates in the saliva, suggests that any such lag is small compared with the 12-hour sampling period.

Experimental Methods

The methods used were as described in Part I, except that, in addition, the phosphorus intake of Sheep 3 and 4 was measured by weighing and analysing not only the bulk supply, but also the residues of food uneaten by the animals. This was necessary because of variations in the composition of the residue from day to day.

Results

Table 6 shows the results of calculations by the above equation for Sheep 3 and 4. Columns 2, 4, 7 and 10 are measurements made at

TABLE 6. Absorption of Dietary Phosphorus

	(1) Days after injection ^{32}P	(2) Ingested P	(3) Faecal P	(4) Urinary P	(5) Total excreted P	(6) Phosphorous Balance	(7) Endog. Faecal P	(8) Dietary P absorbed	(9) % P absorbed	(10) Ca/P of food ingested
		g.	g.	g.	g.	g.	g.	g.		
	2	6.6	6.3	0.2	6.5	0.1	3.2	3.5	53	2.4
Sheep 3	3	6.6	7.3	0.2	7.5	—0.9	2.8	2.1	32	2.4
	4	6.4	4.2	0.3	4.5	1.9	2.6	4.8	75	2.4
	6	5.6	4.7	0.3	5.0	0.6	2.3	3.2	57	2.1
$\Delta t = 24$ hrs.	7	6.0	3.8	0.3	4.1	1.9	2.5	4.7	78	2.3
	8	5.3	4.5	0.3	4.8	0.5	1.7	2.5	47	2.2
	9	4.9	4.6	0.3	4.9	0.0	2.2	2.5	51	2.4
Total for Seven Days		41.4			37.3		17.1	23.3	56	
	4	5.2	2.8	0.01		2.4	2.6	5.0	96	2.0
	5	5.6	2.4	0.01		3.2	2.4	5.6	100	2.1
	6	5.4	1.8	0.01		3.6	1.9	5.5	102	2.0
	7	5.1	2.3	0.01		2.8	1.8	4.6	90	2.2
	8	5.8	1.9	0.01	As	3.9	1.3	5.2	90	2.3
Sheep 4	10	6.1	2.9	0.01	Col.	3.2	1.6	4.8	79	2.3
	11	4.5	3.2	0.01	(3)	1.3	1.9	3.2	71	2.4
$\Delta t = 12$ hrs.	12	5.4	3.4	0.02		2.0	1.7	3.7	69	2.4
	13	6.3	3.8	0.03		2.5	2.0	4.5	71	2.5
	14	6.5	3.6	0.02		2.9	1.9	4.8	74	2.4
	15	6.5	4.5	0.02		2.0	2.1	4.1	63	2.4
Total 1st 6 days		33.2			14.1		11.6	30.7	92	
Total last 5 days		29.2			18.5		9.6	20.3	70	
Total 11 days		62.4			32.6		21.2	51.0	82	

time t (Col. 1), while the figures in Col. 3 were obtained at time t+ Δt. Column 5 = Cols. 3 + 4; Column 6 = 2 — 5; Column 7 = $f\left(\frac{t + \Delta t}{\alpha(t)}\right)$; Column 8 = 2— (3 — 7); and Column 9 is 8/2 × 100. Column 10 is the actual daily calcium–phosphorus ratio of the food consumed. The slight variation from day to day is due to the variation in the relative amount of chaff and nuts left uneaten.

DISCUSSION

An inspection of Table 6 suggests that, for Sheep 4, the figures for phosphorus absorption, balance, and percentage absorbed, fall into two groups, the first of six members and the second of five. As far as percentage absorption is concerned the ranges of the two groups do not overlap and this together with an analysis of variance for absorption and balance, as shown in Table 7, indicates that there is little chance of these groups coming from the same population figures.

TABLE 7. ANALYSIS OF VARIANCE OF TWO GROUPS FOR SHEEP 4

Source of Variation	Degrees of Freedom	P Balance		Absorption	
		Sum of Squares	Mean Square	Sum of Squares	Mean Square
Total	10	5.87		5.43	
Groups	1	2.97	2.97	3.05	3.05
Individuals	9	2.90	0.32	1.38	0.15
	F = 9.3			F = 20.3	

(F at 1% level of probability = 10.6)

Hence the figures for this sheep have been treated in two groups, though similar conclusions would be obtained by combining them into one group.

The collective results for Sheep 3 (56%) fall somewhat lower than those reported by Lofgreen and Kleiber (1954) who found an availability of 90% in lucerne hay. However, the earlier work of Kleiber *et al.* (1951) with cows, gave figures of 50% and 64% for lucerne and concentrates, and these figures, taken with the later results for sheep, cover a similar range to the present results. Since the figures for daily absorption vary from 32% to 102%, it is concluded that all the phosphorus of the diet is available, but the absorption is controlled by other factors. To ascertain whether any light can be thrown on these factors, correlations between various pairs of metabolic rates have been calculated and are presented in Table 8.

The only significant correlations are those of uptake (expressed directly or as a percentage) and balance, the more important one being the former. The high correlation co-efficient, which is associated with regression co-efficients of 0.86 and 0.88, indicates that the phosphorus

balance depends directly on the amount of phosphorus absorbed from the diet rather than on the amount excreted.

TABLE 8. CORRELATIONS BETWEEN VARIOUS RATES OF INTAKE AND EXCRETION

x	y	r within groups
Phosphorus balance	% Phosphorus uptake	0.86**
do.	Absorbed phosphorus	0.87**
do.	Endog. faecal phosphorus	—0.20
Absorbed phosphorus	do.	0.25
do.	Ca/P	—0.01
Dietary phosphorus	Absorbed phosphorus	0.46
do.	Endog. faecal phosphorus	0.34
	Sig. diff. at 5%*	0.48
	at 1%**	0.61

The question whether this regression is the same for different sheep can hardly be answered satisfactorily by such a small sample, but Table 9 shows that the mean square associated with differences between sheep is considerably less than that associated with individual errors. This is a strong indication that the relationship is the same for different sheep, which, of course, is only to be expected if an increase in absorption leads to an equal increase in balance.

TABLE 9. ANALYSIS OF ERROR OF ESTIMATE OF REGRESSION OF PHOSPHORUS BALANCE ON PHOSPHORUS ABSORBED

Source of Variation	Sum of Squares	Degrees of Freedom	Mean Square
Within groups	2.23	14	
From individual regression	2.09	12	0.17
Among group regressions	0.14	2	0.07

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GRASSLAND AND SHRUBLAND ON MAUNGATUA, OTAGO

By A. F. Mark, Botany Department, Otago University, Dunedin

(Received for publication, 22 July 1955)

Summary

Upon the summit of Maungatua (2,944 ft.) cushion herb-moor and *Dracophyllum* scrub may be natural climaxes. The rest of the upland area appears climatically suited to silver beech, since it has an average rainfall of 40 in. and evaporation rates comparable with those where beech is persisting and regenerating. The existing shrubland and grasslands are, accordingly, considered to be disclimaxes associated with burning and grazing. In pre-European times burning alone induced dominance by snow tussock. Latterly, where heavy grazing by stock has followed fire, *Cassinia–Hebe* scrub or fescue tussock have replaced snow tussock. Heavy grazing occurs where small patches are burnt, since stock concentrates upon the burns. It also occurs along fence lines.

The composition of communities modified in different degrees by fire and grazing is illustrated by line-interception diagrams, and a vegetation map and floristic list for the area are given.

Introduction

This paper presents results of an ecological survey of grassland and shrubland on Maungatua. This was part of a more extensive study of the vegetation of the range, of which a fuller description has been deposited in the library of the University of Otago (Mark, 1954). It also formed part of a general survey of vegetation and climate in the Dunedin area, the main results of which are being published (Wardle and Mark, in press).

Maungatua is situated 17 miles west of Dunedin City and 10 miles from the coast, on the western margin of the Taieri Plain. It is some 9 miles long and 4 to 5 miles wide, with the axis running south-west to north-east. On the north-western and northern sides gentle slopes lead up to the summit, but above the Taieri Plain (200 ft.) in the south-east, and Waipori Gorge in the south-west, the slopes rise steeply. The summit is a flattened ridge, 2,500 ft. at either end and rising to 2,944 ft. in the centre. The range consists entirely of Paleozoic mica schist.

Log remains and the little hillocks and corresponding hollows left by uprooted trees provide convincing evidence that at some time prior to European settlement the range was covered in forest. At the time of settlement, however, forest had been reduced to its present limits. It is almost confined to the south-east and south-west slopes where it extends in gullies to an altitude of approximately 2,200 ft. but on the faces is mainly below 1,800 ft.

The early settlers grazed Merino sheep and cattle on the slopes (Shaw and Farrant, 1949), but grazing of cattle has since been much restricted, and Merino sheep have been replaced by Romneys. Rabbits were numerous on the north-west slope before being exterminated about 1910 and are still present near the Taieri Plain but in decreasing numbers. Pigs were present in the early days (Shaw and Farrant, 1949) but have since been exterminated. Deer have occasionally been seen.

Records of rainfall and evaporation indicate that a forest climate exists over the whole area studied (Table 1; see also Wardle and Mark, in press), since they show that the surviving beech forest on the seaward face does not in general occupy country that has either a higher rainfall or a lower evaporation rate than the rest of the range. As this forest is regenerating well and tending to advance marginally, the whole range would appear to be within the climatic tolerance of silver beech unless the temperature at the summit is too low. An attempt is being made to test this with transplants to the summit ridge. With this reservation, the grasslands and shrublands which now cover most of the range may be termed disclimax communities (Clements, 1936). They are essentially of pre-European origin and perhaps owe some of their present diversity to not being completely in harmony with existing climatic conditions before the impact of European pastoral practices. A discussion of the climatic data from grassland and shrubland communities follows the description of the vegetation.

Line Interception Method of Vegetation Analysis

The vegetation was analysed by the line interception method (Canfield, 1941) which consists of recording for each species the length over which it is in contact with a line run at an appropriate height through the community. Canfield has pointed out that the method is suited to the analysis of grassland. Its validity was tested by conducting three trial interceptions in a small area of snow-tussock grassland. The results of these were very similar. Accordingly, two interceptions were made with each analysis. For each, a 50-ft. tape was laid at random. Upper-stratum species were recorded over the whole length of the tape at the height where coverage was most complete, and lower-stratum species for the first 2 ft. in every 10 ft. Values for the two line interceptions have been added. Thus the upper-stratum species were recorded over a total length of 100 ft. and ground-stratum species over 20 ft. In the upper stratum, all vegetation vertically above the inch-wide tape was recorded, whereas in the ground-stratum only those plants situated in a $\frac{1}{4}$-in. strip to one side of the tape were measured. In the diagrammatic presentation of the results (Fig. 2) it has been necessary to combine many of the ground-stratum species, but these may be identified from the list of numbers which correspond to those in the species list at the end of this paper. Where a minor species occupied 1% or more of the ground, the percentage follows in brackets.

Structure and Distribution of Grassland and Shrubland Communities

Snow-tussock Grassland (*Tall-tussock Grassland*)

Snow-tussock grassland is dominated by *Danthonia flavescens*. This is the "low-altitude snow-tussock" grassland of the Tussock Grassland Research Committee (1954). It occupies most of the land above approximately 1,800 ft. and in the least disturbed areas forms a dense cover. But dense stands now occur to only a limited extent, since burning and grazing have considerably modified them. The communities on southern slopes differ from those on the sunny slopes in having a relatively high proportion of co-dominant shrubs, chiefly *Cassinia* spp., *Hebe buxifolia*, *Dracophyllum longifolium* and *Coprosma* spp. *Celmisia*

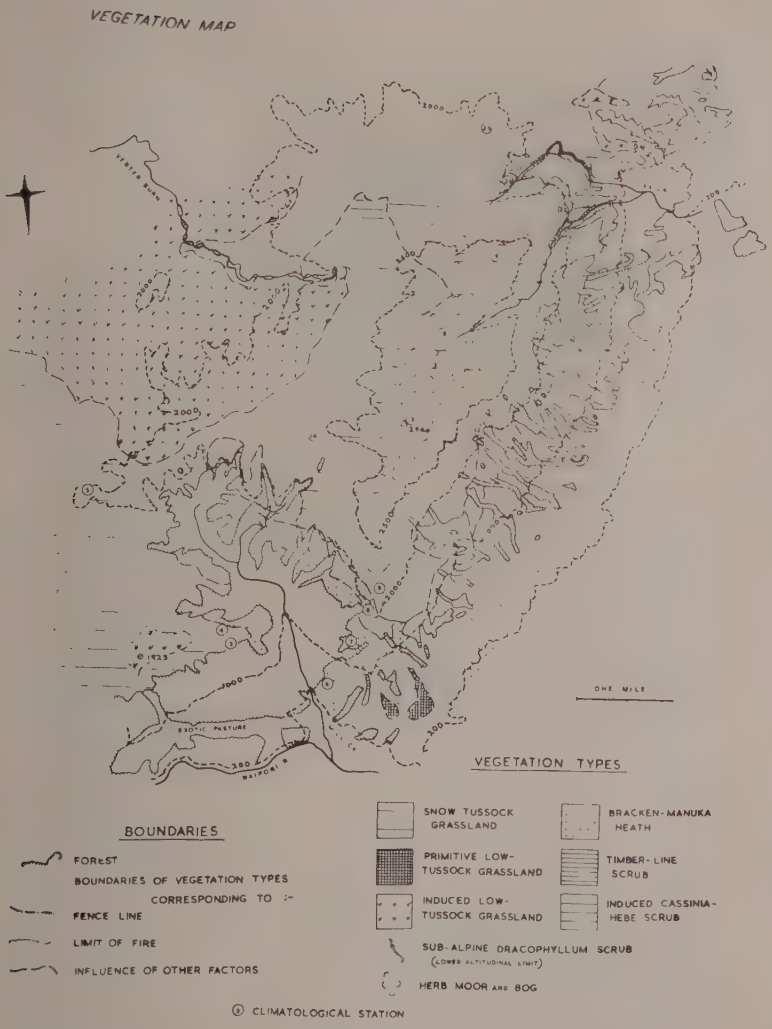

Fig. 1.—Vegetation map of Maungatua.

coriacea var. *stricta* occurs also at higher altitudes. Where the canopy of grass and shrubs is well developed there is a continuous ground cover of which leaf litter forms a substantial part, but the moss *Hypnum cupressiforme, Pernettya macrostigma,* and the small grasses, *Agrostis tenuis, Anthoxanthum odoratum, Poa colensoi, Triodia pumila,* and *Deyeuxia* spp. are also plentiful. This grassland differs from that described for the Hunters Hills, in that co-dominant shrubs are there absent and *Celmisia spectabilis* is abundant in the upper stratum (Barker, 1953).

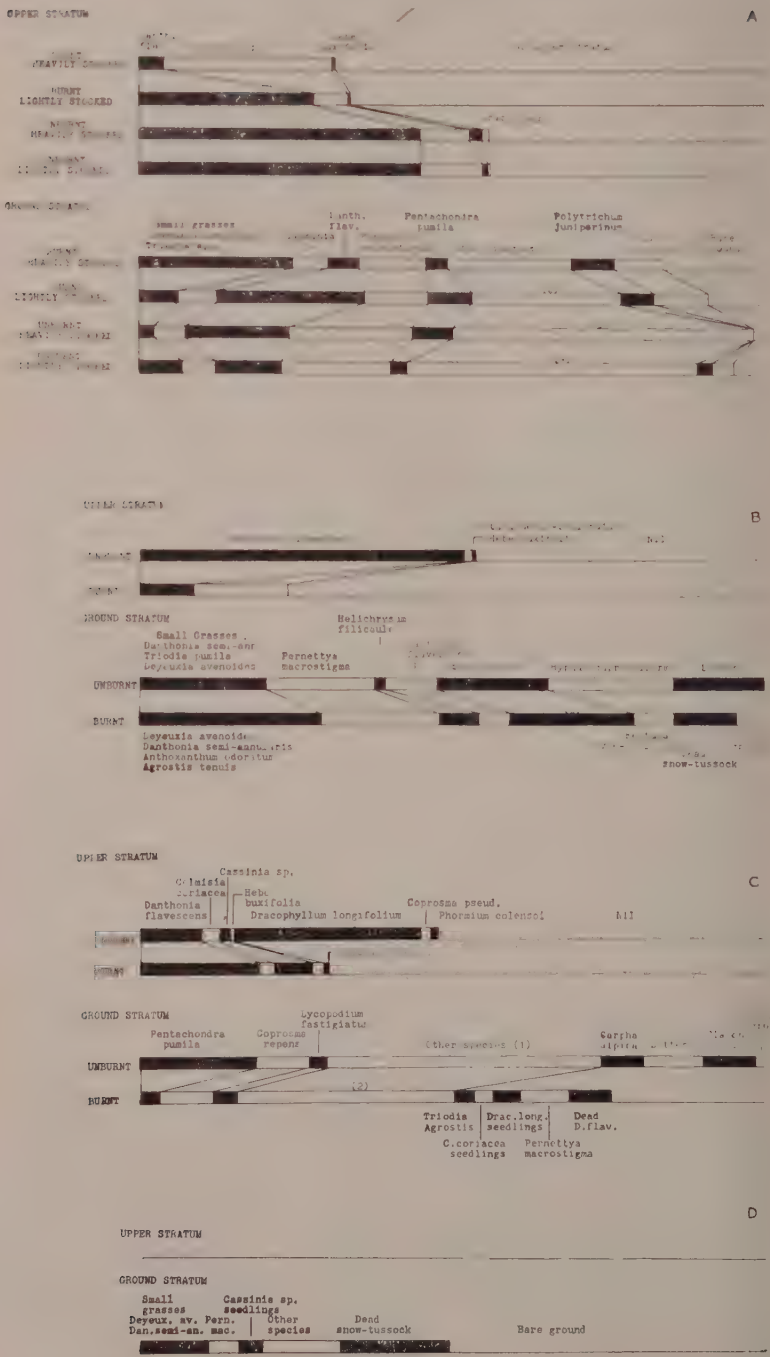
A
Small grasses
Anth. flav.
Pentachondra pumila
Polytrichum juniperinum
B
Helichrysum filicoule
Small Grasses
Danthonia sem.-ar
Triodia pumila
Leycuxia avenoides
Fernettya macrostigma
UNBURNT
BURNT
Leycuxia avenoides
Danthonia semi-aanularis
Anthoxanthum odoratum
Agrostis tenuis
Snow-tussock
C
Cassinia sp.
Hebe buxifolia
Danthonia flavescens
Dracophyllum longifolium
Coprosma pseud.
Phormium colensoi
Lycopodium fastigiatu
Pentachondra pumila
Coprosma repens
Gaultheria
Triodia
Agrostis
Drac.long. seedlings
Dead D.flav.
C.corriacea seedlings
Fernettya macrostigma
D
UPPER STRATUM
GROUND STRATUM
Small grasses
Deyeux. av. Dan.,semi-an.
Cassinia sp. seedlings
Fern.
Mac.
Other species
Dead snow-tussock
Bare ground

Fig. 2.—*Line interception diagrams.**

A. Result of burning six years ago with subsequent heavy and light stocking on the snow-tussock community on the western flank of the summit ridge.

Altitude 2,800 ft.; gentle slope; *Aspect* N.W.

Other species (1) 29 (1.0), 38 (3.5), 58†, 63 (3.0), 71, 79, 84, 87, 88, 92, 93, 102, 117 (4.1), 118, 120 (1.7), 128, 171, 185.

„ „ (2) 1, 21, 24 (1.2), 29, 35 (1.5), 38 (1.7), 44, 58†, 63 (1.2), 71, 79, 80, 84, 92, 109 (1.5), 117 (4.1), 118, 128, 141, 161, (1.0), 175, 185 (1.2).

„ „ (3) 24 (3.0), 29, 35 (4.7), 38 (6.0), 44 (1.2), 58, 60, 63, 71, 79, 80, 84, 88 (1.7), 102, 117 (16.6), 120, 128, 161, 185, (1.5).

„ „ (4) 21, 24 (6.6), 29 (1.0), 35 (1.5), 38 (4.1), 44 (1.8), 48† (2.0), 58, 60, 63, 71 (1.7), 79, 80, 84, 87, 88 (2.2), 93 (1.0), 102, 109, (2.5), 117 (7.1), 118, 120 (1.5), 128, 161, 171, 175, 185 (1.7).

B. Result of burning four years ago on a limited area of pure, dense snow-tussock containing occasional sub-dominant *Festuca novae-zealandiae*.

Altitude, 1,900 ft.; gentle slope; *Aspect* N.W.

Other species (1) 1, 29 (1.2), 63 (2.5), 67, 76, 80 (1.2), 85 (1.7), 93, 101 (3.0), 102 (1.2), 110, 129, 139, 149 (1.2), 150 (2.5), 156 (2.0), 158, 163, 171 (3.0), 175.

„ „ (2) 1 (2.5), 10 (2.5), 29 (2.5), 67, 76, 80, 85 (3.0), 93, 101, 102, 129, 139, 156 (1.2), 158 (2.0), 161, 163, 171 (3.3).

C. Result of burning four years ago on sub-alpine *Dracophyllum* scrub.

Altitude 2,850 ft.; gentle slope; *Aspect* S.W.

Other species (1) 8, 13 (1.2), 27 (1.2), 27†, 29, 31, 35 (1.7), 37 (1.7), 44 (1.2), 48, 58 (2.0), 60 (4.3), 71, 73, 79, 92, 97, 102, 117 (14.2), 123 (1.7). 150, 154, 156, 159, 179, 188 (5.4).

„ „ (2) 24 (1.7), 25†, 27 (1.7), 29 (1.7), 35 (3.0), 37†, 44 (1.8), 48 (3.2), 60 (1.7), 63, 67, 71, 76, 79, 80 (1.0), 84, 88† (1.2), 92 (1.8), 102, 117, 129 (1.2), 131 (1.7), 150, 156, 159, 161, 179 (3.0), 188 (3.0), 190, 196.

D. Effect of autumn burning three years ago on tall, rank snow-tussock containing co-dominant shrubs, chiefly *Dracophyllum longifolium*.

Altitude 2,300 ft.; steep slope; *Aspect* S.

Other species 2†, 18, 48, 58†, 63, 71, 80, 88†(1.7), 92, 101, 102 (3.7), 122, 128, 129, 139, 150, 161, 190 (1.2).

*The numbers refer to the species listed in the appendix. Figures in brackets are percentage of ground covered.

†Denotes seedlings.

Low-tussock Grassland

Two types of low-tussock grassland communities have been recognized:–

1. A primitive low-tussock grassland which occupies small areas below 1,000 ft. on some spurs of the south-east slope. It is dominated by widely spaced tussocks of *Poa caespitosa* and *Festuca novae-zealandiae* with occasional emergent clumps of *Phormium colensoi*. In the continuous ground stratum, *Bulbinella hookeri* and the small grasses *Dichelachne crinita, Anthoxanthum odoratum, Holcus lanatus, Agrostis tenuis, Danthonia semi-annularis,* and *Agropyron scabrum* are abundant. Apart from the exotic grasses, this grassland is probably much as it was at the time of European settlement, although it is now being invaded by manuka (*Leptospermum scoparium*).
2. An induced low-tussock grassland occurring up to 2,000 ft. on the north-west slope. In this community *Festuca novae-zealandiae* is co-dominant with a variable amount of snow-tussock, and is increasing in abundance through the influence of burning and subsequent grazing. Tall shrubs are of negligible occurrence. Individual tussocks of both *Festuca* and *Danthonia* are small, averaging 12 in. in height, and are widely spaced (Fig. 2B). The ground stratum consits of a relatively close sward of *Pernettya macrostigma* and the small grasses listed for snow-tussock grassland, plus *Holcus lanatus*. Only a low proportion of the ground is bare and the community thus has relatively high grazing value.

Sub-alpine Dracophyllum *Scrub*

This sub-alpine scrub is dominated chiefly by *Dracophyllum longifolium*, with the following upper-stratum plants (in decreasing order of abundance): – *Danthonia flavescens, Celmisia coriacea* var. *stricta, Cassinia* spp., *Coprosma pseudocuneata, Phormium colensoi,* and *Hebe buxifolia*. The ground stratum contains a number of typically alpine species, e.g. *Carpha alpina* and *Oreobolus* spp. This community occupies the least-disturbed parts of the summit ridge and descends to 2,200 ft. on poorly drained knolls. Some of the largest *Dracophyllum* shrubs (about 5 ft. high and 4 in. in basal diameter) have approximately 100 growth rings. *Dracophyllum* scrub is regarded by Cockayne as a natural climax in some sub-alpine habitats (1928, pp.273, 280). On Maungatua, the summit-ridge scrub is possibly equivalent to that which normally forms a belt above the timber-line so that its present isolation from the forest indicates that the timber-line is unnaturally depressed.

Induced Cassinia–Hebe *Scrub*

This scrub, dominated by a variable mixture of *Cassinia* spp. and *Hebe buxifolia,* occurs in widely scattered areas above approximately 1,700 ft. In all cases it has been induced through the weakening of other vegetation by burning and grazing. On the summit ridge it appears to have replaced sub-alpine *Dracophyllum* scrub. On the inland (north-west) slope it is restricted chiefly to homeward boundaries of grazing blocks and has replaced snow-tussock grassland. On the seaward slope large areas have originated either from timber-line scrub or from snow-tussock grassland. *Celmisia coriacea* var. *stricta* and *Dracophyllum longifolium* are common members of the community at higher altitudes and on the main south-east face.

Timber-line Scrub

Timber-line scrub occupies small areas close to the upper forest margins on the seaward (south-east) slope. It consists for the most part of shrub and small tree species which provide the under-stories of the adjacent silver-beech forest.

Seral Bracken–Manuka Heath

Heathland, dominated by either the bracken fern (*Pteridium esculentum*) or manuka (*Leptospermum scoparium*) occupies most non-forested areas below 1,600 ft. on the south-east slope, and some small areas of northern aspect up to 2,000 ft. Co-dominant with the bracken, especially along sheltered forest margins, occur *Coriaria sarmentosa* and naturalized *Hypericum androsaemum*, *Leycesteria formosa*, and occasional patches of *Ulex europaeus*. The sparse ground stratum consists for the most part of introduced weed species, e.g. *Rumex acetosella, Hypochaeris radicata* and *Cirsium arvense*.

Cushion Herb-moor

Herb moor occupies poorly drained sites on the summit ridge above approximately 2,750 ft. It consists of turf-forming, prostrate and cushion species; chiefly the cushion plants *Donatia novae-zealandiae, Phyllachne colensoi, Celmisia argentea, Oreobolus pectinatus, O. pumilio, Gaimardia setacea,* and *G. pallida;* the mat shrubs *Dracophyllum prostratum, Pentachondra pumila,* and *Cyathodes pumila;* the sundew *Drosera arcturi,* and lichens belonging to the genera *Cladonia* and *Thamnolia*.

Bog

Although *Sphagnum* species are rare in true herb-moor, typical *Sphagnum* bog is developed to a limited extent on the summit ridge. Of the accompanying species, the most numerous are tussocks of *Phormium colensoi* and *Danthonia flavescens,* shrubs of *Hebe buxifolia, Dracophyllum longifolium,* and *Dacrydium bidwillii,* and smaller plants, especially the sedges *Carex ternaria* and *Carpha alpina*.

EFFECTS OF MANAGEMENT PRACTICES

Snow-tussock Grassland

None of the shrubs or larger grasses are normally browsed to any extent by stock, whose presence, therefore, does not in itself significantly alter the dominant species (Fig. 2A). Furthermore, the tussocks make new foliage rapidly after burning, so that fire alone has little effect upon them except to relieve them temporarily of competition from shrubs. But burning areas to which stock have access can drastically affect the vigour of the tussock since the new growth produced after fire is highly palatable. In short, burning and grazing, while separately producing little permanent change, together can greatly alter the vegetation. Two different management programmes are adopted over the area and this has resulted in two distinct vegetation types.

An almost pure snow-tussock community with a negligible number of tall shrubs has been produced where stocking has been light and burning carried out at approximately six-yearly intervals when most

of a 6,500 acre block is swept by an early-spring (mid-August) fire. Since there is a wide burnt area for the flock to disperse upon, subsequent grazing does not prevent recovery of the old tussocks together with some establishment of seedlings. Six-year intervals between burnings are perhaps over long as the accumulation of litter produces a severe fire which appears to retard recovery of both tussock and subordinate vegetation. Moreover, in that time individual tussocks become large and their shade favours a ground stratum of mosses (*Hypnum cupressiforme*) and *Pernettya macrostigma* rather than small grasses. These features are shown in a line interception of snow-tussock grassland unburnt for 6 years (Fig. 2A). The practice that best maintains the vigour and dominance of snow-tussock while avoiding development of an excessively dense vegetation thus appears to be widespread burning in spring at intervals of 3 or 4 years. Fires at longer intervals, or in the autumn (Fig. 2D), may be sufficiently hot to weaken or kill the tussocks.

A very different result has been produced where relatively small areas are burnt annually. Indiscriminately lighted fires usually fail to run any distance and land managed in this way develops into a mosaic of seral communities with sharp changes in composition at the limits of each burn, or where fences have differentiated the intensity of grazing during the critical period of tussock regrowth (Figs. 3 and 4). Sheep from a wide area congregate upon the recently burnt patches, browsing the new foliage of the tussocks and so weakening them that many fail to recover. This has led to reduction in the cover afforded by snow tussock and in some cases almost complete elimination of this species. Temporarily, there may be a gain in the area occupied by the small grasses, *Deyeuxia avenoides, Danthonia semi-annularis, D. pilosa,*

Fig. 3.—Three communities, bounded by fence lines which differentiate grazing intensity. Snow-tussock retained under light grazing (right background). Low-tussock grassland induced by heavy grazing (foreground). *Cassinia–Hebe* scrub induced by extreme overgrazing (left background); this scrub is now incapable of carrying fire.

Fig. 4.—Induced snow-tussock grassland on western flank of summit ridge burnt six years ago. *Cassinia-Hebe* scrub predominates on area to right of fence line which, being a homeward boundary, is heavily stocked by sheep. Light grazing to left of fence line has permitted recovery of snow tussock. The line interceptions in Fig. 1 A. were adjacent to this site.

Anthoxanthum odoratum, *Agrostis tenuis*, and *Triodia* spp., whose establishment, together with seedlings of unpalatable species, prevents marked erosion. Trampling by heavy stocking can delay the progress of such of the latter as have a creeping habit—namely *Leucopogon fraseri*, *Helichrysum filicaule*, *H. bellidoides*, *Pernettya macrostigma*, *Pentachondra pumila*, and *Gaultheria rupestris*, but it does not arrest invasion by *Cassinia* spp. and *Hebe buxifolia* which will be far advanced before there is sufficient dry vegetation on the area to carry another fire—indeed scrub that consists mostly of *Hebe* appears to be indefinitely fire-resistant. *Cassinia* is particularly favoured by burning through possessing a root stock which survives and makes vigorous new growth. That the extent to which these shrubs replace snow tussock is governed mainly by intensity of stocking after fire is illustrated in Fig. 3 which shows a dense scrub developed where stock congregate along the home boundary of a large grazing block.

A tendency for hard tussock (*Festuca novae-zealandiae*) to replace the snow tussock under pastoral conditions on northern slopes has been noted for the Hunters Hills (Barker, 1953). A comparable situation exists on the north-west slope of Maungatua, mainly below 2,000 ft. Where restricted areas have been burnt and subjected to heavy grazing, hard tussock has become the sole dominant; the more palatable new growth of the snow tussock is closely grazed and it has become a subdominant species in the process of extinction. Frequent burning and continual stocking maintains a very open upper stratum, but the ground stratum retains an abundance of small grasses, especially *Anthoxanthum odoratum* (Figs. 2B, 3).

Barker (1953, p.43) also noted that burning of snow tussock induced prolific flowering of this species eighteen months later. Observations on Maungatua showed that plants burnt in mid-August of 1953 were flowering freely by the following December, whereas unburnt snow tussock began flowering about one month later.

Low-tussock Grassland

Primitive low-tussock grassland has been severely damaged by burning and the grazing of both sheep and rabbits. This is probably the reason why the grassland is in the final stages of replacement by manuka (*Leptospermum scoparium*) scrub.

Sub-alpine Dracophyllum *Scrub*

Dracophyllum is inflammable and is temporarily eliminated by fire whereas tussocks of *Danthonia* and *Celmisia* and rootstocks of *Cassinia* may survive, and together with *Hebe* they usually increase at once from seed (Fig. 5). Line interceptions (Fig. 2C) show that burning also induces considerable changes in the ground stratum. Leaf litter is destroyed, leaving considerable areas bare. Colonization of these patches by *Pernettya macrostigma*, the moss *Polytrichum juniperinum*, and the small grasses *Triodia* spp., *Deyeuxia avenoides*, *Danthonia pilosa*, and *Agrostis subulata* follows. *Dracophyllum* seedlings are not long in invading the community in numbers that, failing further fires, would restore its dominance and sometimes partially do so. But frequent fires are the rule, and they have converted much of the summit into almost pure snow tussock, a community which they serve to maintain despite the continual appearance of *Dracophyllum* and other shrub seedlings.

Fig. 5.—Effect of burning on sub-alpine *Dracophyllum* scrub. *D. longifolium* has been replaced by an open mixture of smaller shrubs, chiefly *Cassinia* and *Hebe buxifolia*, and *Danthonia flavescens*. Dead *Dracophyllum* shrubs are scattered through the burnt area.

The recovery and multiplication of the tussock after fires indicates that the summit ridge is little frequented by stock.

Cassinia–Hebe *Scrub*

The induction of these shrublands by burning and grazing has already been discussed. Since snow-tussock seedlings can establish themselves in low shrubland, and since strongly growing tussocks appear to weaken and suppress *Cassinia*, there is little doubt that if stock were removed the snow-tussock would substantially regain its dominance at altitudes where *Dracophyllum* is not a competitor. Occasional fires should accelerate the process.

Timber-line Scrub

Modifications produced by burning, include an increased abundance of *Cassinia* spp., *Hebe buxifolia, Phormium colensoi, Cyathea colensoi,* and *Celmisia coriacea* var. *stricta*. A typical *Cassinia–Hebe* scrub has been produced in some cases.

Seral Bracken–Manuka Heath

Bracken appears to be encouraged by burning, whereas manuka is destroyed, except for the well-protected seeds, which are dispersed immediately following a fire. Thus, after burning, bracken becomes dominant while manuka later regenerates under dead parent trees or from the fire margin. In the absence of further burning, manuka eventually over-tops the bracken and appears to be a seral community preceding forest, since forest species become established beneath it. However, succession is usually altogether prevented since run owners clear the heathland by frequent burning in order that sheep may be

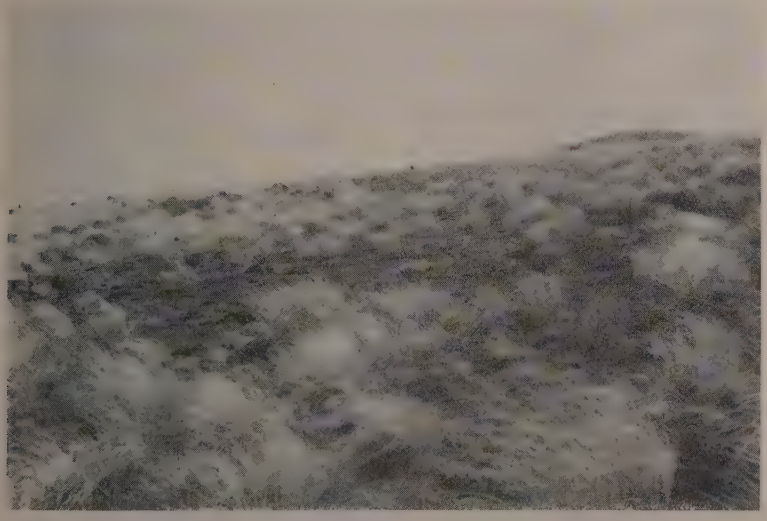

Fig. 6.—Induced snow-tussock community on summit ridge apparently undergoing invasion and replacement by cushion herb-moor. The grids marking out one of the permanent quadrats are shown in position.

mustered from the grazing land at higher altitudes. Thus bracken is dominant over most of it and the soil layer has become thin because of the frequent fires and the steepness of the slope.

Herb Moor

In peaty hollows on the summit ridge, herb-moor appears to be a long-established, edaphic climax community. It is avoided by stock and untouched by fire. Nevertheless, these factors affect it indirectly through weakening the surrounding vegetation, providing opportunity for it to extend on to gentle slopes where its recent origin is indicated by presence of mineral soil close beneath it. Two permanent quadrats have been established to gauge its rate of advance.

CLIMATE IN RELATION TO VEGETATION

Wind records taken at Taieri Airport (88 ft.), 10 miles to the east of Maungatua, show that the prevailing winds are south-westerly. Although these are the chief rain-bearing winds, they may also be associated with periods of fine weather; their velocity tends to increase during spring and summer. Northerly winds (N.E.-N.W.) are more characteristic of the summer months when they are usually accompanied by fine weather.

A year's record of the duration and extent of fog on Muangatua showed that: (1) the range was covered for 116 half-days which were fairly evenly distributed throughout the year; (2) the average fog-base tended to lie between 2,000 and 2,500 ft.; (3) widespread rain accompanied fog on 6 out of every 7 half-days; (4) afternoon fogs were slightly more frequent than morning fogs; (5) in comparison with the more coastal Flagstaff and Mt. Cargill, sea fogs were infrequent. Cloud is frequently present a few hundred feet above the summit ridge when skies are clear elsewhere. This must be a factor, in addition to fog, in reducing the intensity of solar radiation.

During 1953, snow fell on 21 days between April and September, whereas at Taieri Airport it fell on 7 days only, between June and September. Records from Taieri Airport showed that the 1953 snowfall did not differ markedly from average. Snow covered the summit ridge for approximately 70 days, but not continuously. On southerly slopes at higher altitudes the total duration of snow cover would be much longer.

Records of monthly maximum and minimum air temperatures from March to June 1953 at altitudes of 600 ft., 1,100 ft., 2,350 ft., and 2,944 ft., on a grassy spur running up to the Trig, showed that the summit ridge is subjected to temperatures consistently below those obtaining at lower altitudes. The June readings (°F.) 3½ ft. about ground level at the 4 stations were:

	Max.	Min.
600 ft.	70	31
1,100 ft.	63	30
2,350 ft.	59	25
2,944 ft.	55	23

A record of monthly ground minima on the summit during the winter of 1954 shows severe ground frosts—May, 8°F.; June, 3°F.; July 12°F. To ascertain whether any temperature difference existed at equivalent altitudes between the seaward and inland aspects of the range, monthly maximum and minimum air readings were taken for a year at stations 12 and 14 which were 1⅔ miles apart on opposite aspects near the Trig. These records did not differ consistently, and never by more than 3°F.

Rainfall and evaporation records for the period April 1953 to March 1954 were taken in an attempt to evaluate the relationship between vegetation and climate. The type of instruments used and the methods of compiling and evaluating the data are described elsewhere (Wardle and Mark, in press). Results are given in Table 1.

TABLE 1. RECORDS OF RAINFALL AND EVAPORATION AT STATIONS LOCATED IN OR ADJACENT TO GRASSLAND AND SHRUBLAND

Station number	Altitude (ft.)	Aspect	Topography	Vegetation	Av. Annual Rainfall (in.)	Evaporation* (litres)
3	1,600	S.E.	Moderate slope	Snow-tussock	36	5.2
4	1,800	—	Hill crest	,, ,,	25	7.5
5	2,000	N.W.	Gentle slope	,, ,,	40	—
9	2,200	S.W.	Gentle slope	,, ,,	32	—
12	2,350	S.E.	Steep slope	,, ,,	42	5.5
13	2,944	—	Flattish summit	,, ,,	45	4.7
14	2,400	N.W.	Flattish spur	,, ,,	39	4.9
16	2,000	S.E.	Steep slope	,, ,,	43	—
17	2,700	N.W.	Steep slope	,, ,,	36	4.4
19	2,700	N.E.	Flattish spur	,, ,,	45	—
22	2,200	N.	Gentle slope	,, ,,	42	—
23	1,800	S.E.	Gully side	,, ,,	46	—
25	2,260	—	Hill crest	Drac. scrub	40	5.1
18	2,750	S.E.	Steep slope	,, ,,	42	3.4
24	1,300	N.E.	Gentle slope	Induced low-tussock	48	—
8	1,900	S.W.	Gentle slope	Timber-line scrub	41	—
10	600	S.E.	Gentle slope	Heathland	—	5.8
11	1,100	S.E.	Flattish spur	,,	45	5.4
15	1,000	S.E.	Moderate slope	,,	39	—
6	500	S.W.	Steep slope	Pasture replacing forest	32	5.4
7	1,300	S.W.	Steep slope	,, ,,	33	—

* These are atmometer readings for a summer period only (20 November 1953 to 10 March 1954.)

DISCUSSION

If the view is accepted that snow-tussock is nowhere a climatic climax, the fact that it occurs on sites ranging in rainfall from 25 to 46 in. and differing in evaporation rate by 70% merely demonstrates its tolerance as an induced dominant. That fire and grazing rather than climate are indeed controlling factors over most of the area is illustrated by the fact that the highest rainfall recorded (48 in.) was on a site from which snow-tussock has locally been displaced by fescue tussock, ordinarily a low-rainfall species.

Over its entire range, snow tussock tends to share dominance with shrubs, particularly on dark faces where they may largely displace it. Accordingly, the main south-east face of the mountain is largely shrubby while the main north-west face is almost pure grassland. Two pairs of stations were established to assess these aspect differences climatologically. (1) Stations 12 and 14 were on opposite sides of the Trig, the former on the south-east face of the range, the latter in an equivalent position on the north-west face. The second station received three inches less rain than the first but its evaporation rate proved the smaller by 11%. As already stated, there was no consistent temperature difference between these stations. (2) Stations 17 and 18 were about 400 yd. apart on opposite sides of the deep gully that runs north-eastward from near the Trig. They differed in rainfall by six inches and in evaporation by 29%, the scrub-covered dark face being in both respects the moister. No temperature records were kept, but snow was observed to lie much longer on the wetter face.

Probably the climatic climax of the whole area studied is a woody vegetation (mainly beech but possibly replaced by *Dracophyllum* scrub upon the ridge crest), the snow-tussock is a fire climax, and the aspect differences in the present cover merely reflect the lower incidence of fire on dark faces. In part, this is because dark faces are wetter (e.g. station 18 v. 17), but in respect of the upper slopes of the main north-west and south-east faces of the range this difference, if any, is small; indeed the three stations furthest from the coast, 5, 14, and 25, agree closely in rainfall and evaporation, and suggest that in general the country above 2,000 ft. that lies to the north and west of the summit ridge is no drier than that to the east and south. Accordingly, comparative freedom of the former area from shrubs seems most likely to depend upon burning having been more frequent in the days before the advent of stock when fire greatly favoured tussock. These fires, mainly of Maori origin, would originate readily in the large area of drier low-tussock grassland to the west with which it is continuous. This explanation of course assumes that the present uniformity of climate above the 2,000 ft. contour has prevailed for some time.

The low-tussock grassland barely crosses the 2,000 ft. contour. It is not thereby confined to the sites of lower rainfall—for both the wettest (24) and driest (4) of the stations were between 1,000 and 2,000 ft. and low-tussock was present at the former and absent from the latter. The temperature lapse-rate with altitude would seem to determine its upper limit, and fire and grazing to determine its presence or absence within its climatic zone. Bracken–manuka heath has also been induced only below 2,000 ft.

The vegetation of the summit ridge is not under a particularly high rainfall (average 45 in.) but drainage is impeded, and evaporation rates and temperatures are lower than elsewhere. All these factors appear to be necessary for development of herb moor which occurs only on the summit. However, patches of *Dracophyllum* scrub occupy flat ridge-crests down to 2,200 ft. (station 25), but always on shallow, peaty, ill-drained soil which contrasts with the deep clay loam under the surrounding tussock.

The only published meteorological records from similar country are those of Barker (1953) for Hunters Hills. There the rainfall appears to be slightly higher than on Maungatua and the temperatures more extreme. Thus from Barker's data an average annual rainfall of approximately 50 in. can be calculated for the station at 3,000 ft. in snow-tussock, 5 in. more than the summit rainfall of Maungatua. However, the calculated average for the low-tussock station at 1,500 ft. is 37 in. which is within the range encountered about this altitude on Maungatua. Extreme maximum and minimum air temperatures recorded on the Hunter Hills from August 1947 to July 1948 were 97°F. and 17°F. at the 1,500 ft. station, and 92°F. and 13°F. at 3,000 ft. Extremes recorded at 2,400 ft. on the north-west slope of Maungatua (station 14) from July 1953 to June 1954 were only 84°F. and 22°F. Evaporation values cannot be directly compared since the instruments were quite dissimilar, but reduction of evaporation towards the summit is a feature of both ranges.

The vegetational differences between Maungatua and Hunters Hills appear to be considerable. The Hunters Hills have no herb moor, and though some of the Maungatua shrubs are present they do not tend to be co-dominant with, let alone displace, the tussock grasses. Some of the species important in maintaining ground cover between weakened tussocks on Maungatua (notably *Pernettya macrostigma, Herpolirion novae-zealandiae,* and *Triodia australis*) are absent, and soil may remain bare long enough to become conspicuously eroded. *Celmisia spectabilis* takes the place of *Cassinia* as the species most favoured by burning and grazing. These differences scarcely seem explicable on the basis of rainfall and temperature differences alone. It is probable that the evaporation rate on Hunters Hills is higher. Moreover, the soils are derived not from schist but from greywacke.

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APPENDIX

Plants Recorded from Grassland and Shrubland of Maungatua

Nomenclature of vascular plants follows Cheeseman (1925), except where an authority for the name is cited. Only the most conspicuous mosses and lichens have been included, and Mr. William Martin has kindly assisted in their identification.

Species occurring on Maungatua but absent from the Hunters Hills (Barker, 1953) are marked with an asterisk. The letters in parenthesis after species names denote the communities in which they occur in accordance with the following legend:—

Community	*Symbol*
Bracken manuka heath	H
Primitive low-tussock grassland	Ta
Induced low-tussock grassland	Tb
Snow-tussock grassland	Tc
Timber-line scrub	S
Sub-alpine Dracophyllum scrub	D
Herb moor	M
Bog	B

Spermatophyta

Indigenous species

1 *Acaena sanguisorbae*: H, Ta, Tb, Tc.
2 *Aciphylla colensoi*: Tb, Tc.
*3 *A. scott-thomsoni* Cockayne: S.
4 *Agropyron scabrum*: Ta, Tc.
5 *Agrostis dyeri*: Tc.
6 *A. muscosa*: Tc.
*7 *A. parviflora*: Tc, D.
8 *A. subulata*: Tc, D.
9 *Angelica montana*: Tc.
10 *Anisotome aromatica*: Tb, Tc.
11 *Arundo conspicua*: Tc.
*12 *Asperula perpusilla*: Tc.
*13 *Astelia cockaynei*: S, D.
*14 *A. linearis*: D.
15 *Bulbinella hookeri* var. *angustifolia*: H, Ta.
*16 *Caladenia bifolia*: Tc, D.
17 *C. lyallii*: Tc, D.
18 *Carex breviculmis*: Tc, D.
*19 *C. colensoi*: Tc.
*20 *C. gaudichaudiana*: Tc.
21 *C. ternaria*: Tc, D, B.
*22 *C. secta*: Tc.
23 *Carmichaelia subulata*: S.
*24 *Carpha alpina*: D, M, B.
†25 *Cassinia* spp.: Tc, S, D.
*26 *Celmisia argentea*: D, M.

† For a note on the forms of this genus upon Maungatua see Cockayne and Allan (1927).

27 *C. coriacea* var. *stricta*: Tc, S, D.
28 *C. densiflora*: Tc.
29 *C. gracilenta*: Tb, Tc, D.
*30 *C. linearis*: M.
31 *C. lyallii*: D.
32 *C. prorepens*: Tc.
*33 *C. sessiliflora* var. *minor*: M.
*34 *Chiloglottis cornuta*: D.
35 *Coprosma cheesemanii* Oliver: Tc, D.
36 *C. parviflora*: Tc, S.
37 *C. pseudocuneata* Oliver: D.
38 *C. repens*: D, M, B.
*39 *C. rugosa*: Tc, S.
*40 *Cordyline australis*: H.
41 *Coriaria sarmentosa* Forst.: H.
42 *Craspedia uniflora*: Tc.
43 *Cyathodes colensoi*: Tc.
*44 *C. empetrifolia*: Tc, D, S.
45 *C. pumila*: D, M.
*46 *Dacrydium bidwillii*: D, B.
*47 *Danthonia cunninghamii*: S.
48 *D. flavescens* Hook. f.: Tb, Tc, S, D, B.
49 *D. pilosa*: Ta, Tb, Tc.
*50 *D. semi-annularis*: Ta, Tb, Tc.
*51 *Deschampsia novae-zealandiae*: Tc.
52 *Deyeuxia avenoides*: Tb, Tc, S, D.
53 *D. forsteri*: Tc, D.
*54 *D. setifolia*: Tc, S.
55 *D. youngii*: Tc, S.
56 *Dichelachne crinita*: Ta, Tb, Tc, H.
*57 *Donatia novae-zealandiae*: M.
58 *Dracophyllum longifolium*: Tc, D, S, B.
*59 *D. politum*: D, M, B.
*60 *D. prostratum*: D, M, B.
*61 *D. uniflorum*: D.
*62 *Dracophyllum* hybrid forms: D, M, B.
63 *Drapetes dieffenbachii*: Tb, Tc, D.
*64 *Drosera arcturi*: M, B.
65 *Echinopogon ovatus*: Tc.
*66 *Epilobium rotundifolium*: S.
*67 *E. tenuipes*: Tb, Tc, S, D.
*68 *Euphrasia dyeri*: M, B.
69 *E. zealandica*: Tc, D, M, B.
70 *Festuca novae-zealandiae*: Ta, Tb.
71 *Forstera tenella*: Tc, D, M, B.
*72 *Gaimardia pallida*: M, B.
*73 *G. setacea*: D, M, B.
74 *Gaultheria antipoda*: S.
75 *G. depressa*: Tc.
76 *G. rupestris* R. Br.: Tb, Tc, D.
77 *Gentiana corymbifera* Kirk: Tc.
78 *G. grisebachii* var. *novae-zealandiae*: Tc.
79 *G. patula*: Tc, D, M.
80 *Geranium microphyllum*: Tb, Tc, D.
81 *Geum leiospermum*: Tc, D.
82 *G. parviflorum*: D.
*83 *Gnaphalium paludosum*: D.
84 *G. traversii*: H, D, M.
*85 *Gunnera monoica*: H, Tb, Tc.
*86 *G. strigosa*: Tc.
87 *Haloragis depressa*: Tb, Tc, D.
88 *Hebe buxifolia* (Benth.) Ckn. et Allan var. *odora* Kirk: Tc, D, S, B.
*89 *H. propinqua* (Cheesem.) Ckn. et Allan: Tc, D.
90 *H. salicifolia* (Forst. f.) Pennell: S.
91 *Helichrysum bellidioides*: Tc, D.
92 *H. filicaule*: Ta, Tb, Tc, D.
*93 *Herpolirion novae-zealandiae*: Tb, Tc, D.
94 *Hierochloe fraseri*: D, B.
95 *Hydrocotyle* spp.: H.
96 *Hymenanthera dentata* var. *alpina*: Tc, D.
*97 *Juncus antarcticus*: Tc, D.
98 *Koeleria kurtzii*: Tc.
99 *Lagenophora pumila*: H.
*100 *Leptospermum scoparium*: H, Tc.
101 *Leucopogon fraseri*: H, Ta, Tb, Tc.
102 *Luzula campestris*: Tb, Tc, D.
*103 *Lyperanthus antarcticus*: D.
104 *Mazus radicans*: Tc, D.
105 *Mentha cunninghamii*: Tc.
*106 *Microseris scapigera*: Tc.
107 *Microtis unifolia*: Tc, D.
108 *Muehlenbeckia complexa*: D.
109 *Myrsine nummularia* Hook. f.: D.
*110 *Nertera ciliata*: Tc.
*111 *Nothopanax colensoi*: S.
*112 *Olearia arborescens*: S.
*113 *O. cymbifolia*: S.
*114 *O. nummularifolia*: S.
115 *O. virgata*: Tc.
116 *Oreobolus pectinatus*: D, M.
117 *O. pumilio*: D, M.
118 *Oreomyrrhis andicola* var. *colensoi*: Tc, D.
119 *O. a.* var. *ramosa*: Tc, D.
*120 *Oreostylidium subulatum*: Tc, D.
121 *Oxalis lactea*: Tc.
122 *Pentachondra pumila*: Tc, D, M.
*123 *Pernettya macrostigma* Col.: Tb, Tc, S, D, M, B.
124 *Phormium colensoi*: Ta, Tc, S, D, B.
125 *P. tenax* Forst.: H, Ta.
126 *Phyllachne colensoi* var. *haastii*: M.
*127 *Pimelia lyallii*: Tc, D.
128 *P. prostrata*: Tc, D, M.
129 *Plantago brownii*: Tb, Tc, D.
130 *Poa caespitosa*: H, Ta.
131 *P. colensoi*: Tb, Tc, D.
*132 *P. imbecilla*: Tc, D.
*133 *P. lindsayi*: Tc.
*134 *P. pusilla*: Tc, S.

135 *Prasophyllum colensoi*: Tc.
136 *Pratia angulata*: Tc, D, B.
*137 *Pterostylis australis*: Tc.
138 *Ranunculus hirtus*: H.
139 *R. lappaceus* var. *multiscapus*: Tb, Tc.
*140 *R. sinclairii* var. *angustatus*: D, B.
141 *Raoulia glabra*: Tb, Tc, S.
*142 *R. grandiflora*: Tc.
143 *R. subsericea*: Tc.
144 *Rubus cissoides* A. Cunn.: S.
145 *Schoenus pauciflorus*: Tc.
*146 *Scirpus antarcticus*: Tc.
*147 *S. aucklandicus*: Tc, D.
*148 *S. inundatus*: Tc.
149 *Scleranthus biflorus*: Tb, Tc.
150 *Senecio bellidioides*: Tb, Tc, D.
*151 *Stellaria gracilenta*: Tc.
152 *Thelymitra longifolia*: Tc, D.
153 *T. uniflora*: Tc, D.
*154 *Triodia australis*: Tc, D.
155 *T. pumila*: Tc, D.
*156 *Uncinia filiformis*: Tc.
*157 *U. purpurata*: Tc.
*158 *U. rubra* Boott.: Tb, Tc, D.
*159 *U. rupestris*: Tc.
*160 *Utricularia monanthos*: M, B.
161 *Viola cunninghamii*: Tb, Tc, D.
*162 *V. filicaulis*: H.
163 *Wahlenbergia albomarginata*: Tb, Tc, D.

Exotic Species

164 *Agrostis tenuis* Sibth.: H, Ta, Tb, Tc, D.
*165 *Aira praecox* L.: Tc.
166 *A. caryophyllea* L.: Tc.
167 *Anthoxanthum odoratum* L.: H, Ta, Tb, Tc.
*168 *Betula pubescens* Ehrh.: S.
169 *Cirsium arvense* L.: H.
170 *Holcus lanatus* L.: H, Ta, Tb.
171 *Hypochaeris radicata* L.: H, Ta, Tb, Tc, D.
*172 *Hypericum androsaemum* L.: H.
*173 *Leycesteria formosa* Wall: H.
*174 *Pinus radiata* Don.: Ta, D.
175 *Rumex acetosella* L.: H, Tb, Tc, D.
176 *Trifolium repens* L.: H, Ta, Tb, Tc.
177 *Ulex europaeus* L.: H, Tc, S.

Filices

178 *Blechnum minor* (R.Br.) Cockayne: Tc, S, D.
179 *B. penna-marina*: Tc, D.
*180 *Cyathea colensoi* (Hook. f.) Domin.: S.
181 *Hymenophyllum multifidum*: Tc.
182 *Hypolepis millefolium*: Tc.
183 *Ophioglossum coriaceum*: D.
184 *Polystichum vestitum*: Tc, S.

Lycopodiaceae

185 *Lycopodium fastigiatum*: Tc, S, D.
*186 *L. selago*: Tc, D.
*187 *L. volubile*: S.

Musci

188 *Dicranoloma pungentella* (C.M.) Par: D, M.
189 *Hypnum cupressiforme* Hedw.: Tc.
190 *Polytrichum juniperinum* Willd.: Tb, Tc, D.
191 *Rhacomitrium lanuginosum* (Hedw.) Brid. var. *pruninosum* (C.M.) H.f. & W.: D, M.
192 *Sphagnum* spp.: M, B.

Lichenes

*193 *Cladonia alpestris* (L) Rabh. var. *sylvestris*: M.
*194 *C. amaurocrea* Schaer: M.
*195 *C. retipora* Flke.: D, M.
*196 *C. sullivani* Mullarg: M.
*197 *C. uncialis* (L.) Web.: D.
198 *Stereocaulon ramulosum* Ach.: Tc, D.
*199 *Thamnolia vermicularis* Ach.: M.
*200 *Usnea contexta* Mot.: M.

TOXICITY OF SOME HYDROCARBON INSECTICIDES TO EARTHWORMS

By J. M. Hoy, Entomological Research Station, Department of Scientific and Industrial Research, Palmerston North

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Summary

Toxicity of DDT, crude BHC, and gamma BHC, to earthworms in soil was tested at various dosages. In soils of moderate organic-matter content, significant mortality occurred with DDT at 80 lb. p.p.i. per acre. Mortality was not significant with DDT at 40 lb. p.p.i. per acre, gamma BHC at 40 lb. g.i. per acre, or crude BHC at 20 lb. g.i. per acre. In soil of low organic-matter content 21 lb. g.i. crude BHC killed all earthworms.

These three insecticides are unlikely to harm earthworms if applied at the rate of 2 lb. effective isomer per acre as used for the control of *Costelytra zealandica* (White) in pastures.

Introduction

The use of chemical methods for the control of the common grass grub *Costelytra zealandica* (White) in pastures (Kelsey and Hoy 1950) has lead to a rapid increase in the amount of insecticide applied to agricultural land in New Zealand. Large-scale use of insecticide focuses attention on possible deleterious effects on beneficial soil organisms. For this reason the effect of DDT and benzene hexachloride (BHC) on earthworms was investigated.

The literature contains few references to the toxicity of hydrocarbon insecticides to earthworms. Greenwood (1945) reported the death of all earthworms exposed to 100 lb. of DDT per acre for one month. Baker (1946) noted a reduction in earthworm populations following the use of unspecified amounts of 0.25% emulsified DDT as a foliage spray to trees.

Materials and Methods

Laboratory Experiment

Earthworms used were taken from pasture soil, the species being *Lumbricus rubellus* Hoffmeister and *Allolobophora caliginosa* (Savigny), the former being the more numerous. These species are commonly found in pasture soils throughout New Zealand (K. E. Lee pers. comm.).

Experiments were conducted in 4-in. jars using the surface area for calculation of equivalent per-acre dosages of insecticide, which was evenly incorporated with the soil. For lower rates of application, at 2 and 10 lb. per acre equivalent, five time the necessary quantity of insecticide was suspended in one litre of water, 200 cc. being drawn off and applied evenly to sufficient soil for all replicates. Soil was thoroughly mixed and divided on a weight basis. For higher rates of application the insecticide was weighed, bulked with sand, and mixed first with quarter the total bulk of soil before being thoroughly incorporated with the remainder. All soil was partially sterilized to prevent the carryover of earthworms.

In the first series, in which soil of low organic-matter content was used, jars were seeded with ryegrass and held for one week prior to introduction of earthworms. In the second and third series using soil of moderate organic-matter content the jars were not seeded. Soil-moisture level was maintained by addition of water from time to time. All jars were held at room temperatures. Counts were made of earthworms surviving after eight weeks' exposure.

Details of experiments are as follows:—

SERIES I.

Six replications each with six earthworms, soil of low organic-matter content.

Materials Used	Rate per acre (effective isomer, lb.)
DDT 10% p.p.i. dust	21
Crude BHC 10% g.i. wettable powder	21
Control	—

SERIES II

Five replications each with six earthworms, soil of moderate organic-matter content.

Materials Used	Rate per acre (effective isomer, lb.)
DDT 50% p.p.i. wettable powder	40
Crude BHC 10% g.i. wettable powder	2, 10
Gamma BHC 50% g.i. wettable powder	2, 10
Control	—

SERIES III

Replications, numbers, and soil as for series II.

Materials Used	Rate per acre (effective isomer, lb.)
DDT 50% p.p.i. wettable powder	80
Crude BHC 10% g.i. wettable powder	$12\frac{1}{2}$, 15, $17\frac{1}{2}$, 20
Gamma BHC 50% g.i. wettable powder	15, 20, 30, 40
Control	—

Field Experiment

In addition to these laboratory experiments a field area treated with DDT, crude BHC, and gamma BHC, at rates of 2, 30, and 60 lb. effective isomer per acre was sampled to determine the decrease in toxicity of these insecticides to earthworms with the passage of time. The insecticides mixed with superphosphate were broadcast and raked into the top inch of soil. Ten 4-in. cores to a depth of nine inches were taken on each plot and control, 23 and 83 weeks after application of insecticides.

EXPERIMENTAL RESULTS

Series I.

Counts taken at the end of the eight weeks showed mean survival rates of earthworms as follows:—

DDT at 21 lb. p.p.i. per acre	6.0 ± 0.2 per replicate
Crude BHC at 21 lb. g.i. per acre	0.0 „ „
Control—no treatment	5.6 ± 0.2 „ „

Series II and III

Analyses of variance were carried out on data in these series. Data shown in Tables 1 and 2 are counts of survival of the original 6 earthworms per jar.

TABLE 1. SURVIVAL OF EARTHWORMS IN LABORATORY TRIAL—SERIES II—AFTER EXPOSURE FOR EIGHT WEEKS

Material	Active Isomer (lb./ac.)	No. of Live Earthworms
DDT	40	5.6
Crude BHC	2	5.8
	10	5.8
Gamma BHC	2	5.6
	10	5.4
Control	—	5.4
S.E. of means		± 0.26

There were no significant differences due to treatment in Series II. In Series III the mean of the two heaviest gamma BHC treatments was

TABLE 2. SURVIVAL OF EARTHWORMS IN LABORATORY TRIAL—SERIES III—AFTER EXPOSURE FOR EIGHT WEEKS

Material	Active Isomer (lb./ac.)	No. of Live Earthworms
DDT	80	2.0
Crude BHC	12.5	5.4
	15	6.0
	17.5	5.8
	20	5.8
Gamma BHC	15	5.8
	20	6.0
	30	5.0
	40	5.0
Control	—	5.6
S.E. of means		± 0.27
Sig. diff. (5%)		0.78

significantly lower than the mean of the two lighter treatments, $F = 10.95^*$ for 1,40 d.f. This also appeared as a significant downward trend in the means of gamma BHC with increasing rate of application, $F = 8.52^*$ for 1,40 d.f.

Field Plots

Data obtained from field plot sampling can only be analysed on the basis that differences between plots were entirely due to treatments and not to individual variations in earthworm populations. On this basis analyses of variance were made. Earthworm survival in plots 23 weeks, and earthworm recovery 83 weeks after insecticide application —treatment means, their standard errors and the difference rquired for significance at the 5% level of probability are shown in Table 3.

The Material × Time interaction from a combined analysis of variance gave $F = 4.45^*$ (2,180 d.f.). In the 60 weeks between counts the two sets of BHC treatments retained their toxicity while there was a significant drop in toxicity in the DDT treatments over the same period.

TABLE 3. EARTHWORM COUNTS IN PLOTS: 23 AND 83 WEEKS AFTER INSECTICIDE APPLICATION

Material	Active Isomer (lb/ac)	Live Worms at: 23 weeks	83 weeks
Crude BHC	60	0.9	1.3
	30	1.0	1.1
	2	2.7	3.0
Gamma BHC	60	2.5	3.4
	30	2.5	2.6
	2	3.3	3.9
DDT	60	1.0	2.2
	30	1.3	5.2
	2	3.3	5.6
Control	—	3.8	4.9
S.E. of means		± 0.61	± 0.77
Sig. diff. (5%)		1.73	2.17

DISCUSSION

The first series of laboratory experiments indicated the dosage range to be investigated in the second and third series. Soil used in Series I had grown a number of horticultural crops and was relatively low in organic matter. Soil in Series II and III was of the same type but had been ploughed from pasture two years previously and resown to grasses and clovers. It is probable that this difference in organic matter is responsible for the toxic effect of mixed isomer BHC in the first series as compared with the same material in Series III. Similar effects have been noted in relation to phytotoxicity of BHC (Simkover and Schenefelt 1952).

In Series II and III with the insecticide intimately mixed with soil of moderate organic-matter content there was no significant mortality at eight weeks with DDT up to 40 lb. p.p.i. per acre, gamma BHC up to 40 lb. g.i. and crude BHC up to 20 lb. g.i. There was a significant trend of increasing mortality in gamma BHC at dosages above 20 lb. g.i. per acre, crude BHC was not tested in this range.

Conclusions drawn from field plot sampling are, strictly speaking, only valid for the sampling area as no account was taken of initial variations in earthworm population. This area was treated to determine the phytotoxicity of these insecticides to germinating seed and no earthworm sampling was possible until the termination of these trials. An increased level of toxicity was noted in the field plots, this was probably

due to local concentration of insecticide in the surface layer due to method of application. Toxicity of DDT to earthworms was significantly less persistent under field conditions than that of the BHC compounds.

In soils of moderate organic-matter content it would seem that all three insecticides have little effect on earthworms at dosages up to 20 lb. actual toxicant per acre. In the field this amount of insecticide would be applied over a 30-year period for grass-grub control. It would appear that the application of 2 lb. actual toxicant per acre for grub control every third year will not be harmful to earthworm populations.

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SCREENING TESTS WITH FUNGICIDES FOR CONTROL OF CELERY LEAF SPOT (*Septoria apii* Chester)

By H. Jacks, Plant Diseases Division, Department of Scientific and Industrial Research, Auckland

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Summary

Of thirty fungicides tested in the glasshouse for control of *Septoria apii* Chester, those based on ferbam, thiram, zineb, maneb, dichlone, captan, nitrobenzene, dinitro phenyl crotonate, chloranil, lime sulphur, and ziram, were the most effective.

When treated plants were watered to simulate very heavy rain (1 in. in 6 min.), ferbam, zineb and dichlone formulations were more effective than other compounds in preventing leaf spot infection.

Introduction

Leaf spot (*Septoria apii* Chester) of celery (*Apium graveolens* L.) was recorded in New Zealand by Cunningham (1922), and described by Neill (1933), who recommended preventive spraying with Bordeaux (6 : 8 : 100). Tests outlined in this paper were carried out as part of the Division's certification scheme to ascertain the effectiveness of various fungicides in control of this disease.

Materials

Copper compounds: Bordeaux mixture; Cop-ox (50% copper oxychloride) wettable powder; Perenox (50% cuprous oxide) wettable powder; Blue Ammon (4.5% copper ammonium complex) liquid; Ialine colloidal copper (15% copper oxychloride) emulsion; and Fungex (8.5% copper ammonium carbonate) liquid.

Sulphur compounds: Lime sulphur (15% polysulphides) liquid; Colsul 40 (40% colloidal sulphur) paste; and Cosan (75% sulphur) wettable powder.

Dithiocarbamates: Fermspray (70% ferbam) wettable powder; Fertram (80% ferbam) wettable powder; Dithane D14 (x/y/z = x lb. of 19% nabam/y lb. zinc sulphate/z lb. hydrated lime) tank mix formulation; Dithane Z78 (65% zineb) wettable powder; Manzate (70% maneb) wettable powder; Zirospray (70% ziram) wettable powder; Fuclasin Ultra (70% ziram) wettable powder; Tetram (80% thiram) wettable powder; Thiotox (80% thiram) wettable powder; and Thirospray (50% thiram) wettable powder.

Miscellaneous compounds: Phygon XL (50% dichlone) wettable powder; Spergon w.p. (48% chloranil) wettable powder; Shirlan A.G. (25% salicylanilide) paste; Flit 406 (50% captan) wettable powder; Orthocide (50% captan) wettable powder; Nirit (15% dinitro benzene) wettable powder; Bulbosit (5% dinitro benzene) dust; Botrilex (20% pentachloronitro benzene) dust; Folosan (16% tetrachloronitro benzene) dust; Crag 341 C (34% glyodin) liquid; and Karathane (22.5% dinitro (1 methyl heptyl) phenyl crotonate + 2.5% other nitrated derivatives of phenol) wettable powder.

TABLE 1. TRIAL 1. EFFECT OF FUNGICIDES APPLIED AT SEVERAL CONCENTRATIONS ON

Copper Compounds				Sulphur Compounds			
Treatment	Amount (lb.) in 100 gal. water	Lesions per leaf stalk		Treatment	Amount in 100 gal. water	Lesions per leaf stalk	
		Mean No.	Log (x + 1)			Mean No.	Log x 1
Check	—	321.3	2.51	Check	—	233.5	2.37
Bordeaux mixture	10 : 8	64.0	1.81	Lime sulphur	2 gal.	9.1	0.89
„	6 : 8	49.4	1.63	„ „	1 gal.	2.1	0.45
Cop-ox	12	23.9	1.36	„ „	0.75 gal.	0.0	0.00
.	10	46.0	1.57	„ „	0.5 gal.	23.1	1.13
	6	41.0	1.54	Lime sulphur: Colsul 40	0.75 gal. : 2 lb	27.1	1.35
..	5	34.9	1.45				
Perenox	12	94.7	1.87	„ „	0.5 gal. : 2 lb.	53.9	1.74
„	10	44.4	1.62	Colsul 40	10 lb.	64.9	1.81
..	6	121.0	1.75	„	6 lb.	65.3	1.49
..	5	26.7	1.40	„	4 lb.	55.7	1.67
Blue Ammon	3 gal.*	20.9	1.05	„	2 lb.	51.9	1.72
„	2 gal.	7.9	0.88	Cosan	5 lb.	47.4	1.66
„	1.5 gal.	5.4	0.74	„	2.5 lb.	67.3	1.84
..	1 gal.	13.6	1.05				
..	0.5 gal.	14.7	1.08				
„	2	29.7	1.41				
„	1	23.6	1.35				
Ialine colloidal copper	2 gal.	61.0	1.39				
„ „	1 gal.	67.0	1.80				
„ „	0.5 gal.	90.3	1.93				
Fungex	2 gal.	3.3	0.56				
„	1 gal.	6.3	0.69				
„	0.5 gal.	16.0	1.11				
„	0.25 gal	26.7	1.43				

Difference required for significance at 1%: 0.41

* Fungicides at this strength caused necrotic spotting on leaves.

** Plants dusted to maximum retentive capacity.

DEVELOPMENT OF CELERY LEAF SPOT (Means from 3 replications each of 5 plants.)

Dithiocarbamates				Miscellaneous compounds			
Treatment	Amount (lb.) in 100 gal. water	Lesions per leaf stalk		Treatment	Amount (lb.) in 100 gal. water	Lesions per leaf stalk	
		Mean No.	Log (x + 1)			Mean No.	Log (x + 1)
Check	—	260.0	2.42	Check	—	533.3	2.73
Fermspray	4	0.0	0.00	Phygon XL	1	1.0	0.21
,,	2	1.6	0.43	,,	0.5	1.0	0.29
,,	1	6.4	0.76	,,	0.25	0.9	0.19
Tetram	4	0.6	0.17	Spergon w.p.	1	0.0	0.00
,,	2	2.9	0.51	,,	0.5	1.9	0.40
,,	1	14.7	1.03	,,	0.25	0.4	0.11
Dithane D 11	4 : 2 : 1	0.0	0.00	Shirlan A.G.	8	18.7	1.13
,,	2 : 1 : 0.5	1.7	0.38	,,	4	31.9	1.45
Dithane Z 78	4	3.4	0.50	,,	2	42.7	1.56
,,	2	2.1	0.35	Flit 406	4	0.3	0.04
,,	1	25.9	1.35	,,	2	2.0	0.34
Manzate	4	0.6	0.13	,,	1	4.0	0.59
,,	2	5.0	0.40	Nirit	10	1.0	0.27
,,	1	158.9	2.18	,,	7.5	6.1	0.80
Zirospray	4	7.9	0.85	,,	5	1.6	0.39
,,	2	2.0	0.40	,,	2.5	28.6	1.47
,,	1	20.0	1.30	Bulbosit	2.5g.**	324.0	2.13
				Botrilex	1.9g.**	138.9	2.14
				Folosan	1.5g.**	324.6	2.47
				Crag 341C	8*	0.0	0.00
				,,	4*	1.9	0.30
				,,	2	11.6	0.98
				Karathane	2	1.4	0.39
				,,	1	7.6	0.63

EXPERIMENTAL METHODS

Celery seedlings of the variety Golden Self Blanching were transplanted into 4-in. pots, 4 weeks after seed was sown. Four weeks later, when 3 leaf stalks had developed, plants were graded according to their size, and different size groups were equally distributed amongst treatments. Five plants per treatment were sprayed with 100 ml. of fungicidal suspensions. Dusts were applied at maximum amounts plants would retain. Twenty-four hours later plants were inoculated by spraying with an aqueous spore suspension of *S. apii* from infected leaves. The

TABLE 2. TRIAL 2. RESULTS FROM FURTHER TESTS WITH TREATMENTS PREVENTING LEAF SPOT IN TRIAL 1, AND SOME MATERIALS BASED ON THE SAME ACTIVE INGREDIENTS AS EFFECTIVE TREATMENTS

(Means from 5 plants.)

Treatments selected from Table 1				Final selections			
Treatments	Amount (lb.) in 100 gal. water	Lesions per leaf stalk		Treatments	Amount (lb.) in 100 gal. water	Lesions per leaf stalk	
		Mean No.	Log (x + 1)			Mean No.	Log (x + 1)
Check		119.0	2.08	Check	—	293.2	2.47
Lime sulphur	0.75 gal.	2.6	0.47	Fermspray	4	1.4	0.25
Fermspray	4	0.6	0.18	Fertram	3.5	2.4	0.41
Fertram	3.5	0.4	0.09	Tetram	4	13.0	0.95
Tetram	4	0.4	0.12	Thiotox	4	11.6	0.91
Thiotox	4	0.0	0.00	Thirospray	3.2	3.6	0.34
Thirospray	6.4	0.0	0.00	Dithane D 14	2 : 1 : 0.5	1.6	0.27
,,	3.2	0.4	0.12	Manzate	2	2.0	0.31
Dithane D 14	2 : 1 : 0.5	0.0	0.00	Fuclasin Ultra	2	14.2	0.93
Dithane Z 78	2	8.0	0.84	Phygon XL	0.25	5.0	0.42
Manzate	2	0.0	0.00	Spergon w.p.	0.25	6.6	0.93
Zirospray	2	9.2	0.64	Flit 406	2	4.6	0.42
Fuclasin Ultra	2	0.4	0.09	Orthocide	2	4.9	0.65
Phygon XL	0.25	0.2	0.06	Nirit	5	0.6	0.12
Spergon w.p.	0.25	0.2	0.06	Crag 341 C	4	6.4	0.91
Orthocide	2	0.2	0.06	Karathane	2	5.0	0.51
Flit 406	2	1.0	0.20				
Nirit	5	0.4	0.12				
Crag 341C	4*	0.8	0.19				
Karathane	2	1.4	0.23				
Difference required for significance at 1%:			0.39				0.65

*Fungicides at this strength caused necrotic spotting on leaves.

suspension, applied at 100 ml. per five plants, contained approximately 10,000 spores per ml. After inoculation, plants were placed in a humidity cabinet at 91 ± 1% relative humidity and 77.5 ± 2.5°F. for 24 hours. Plants were then moved to a glasshouse held at approximately 85°F.

Lesion counts were made on all leaflets on each stalk 14 to 18 days after inoculation. The mean number of lesions per plant is given in the Tables.

Any material used in the first trial (Table 1) that was not significantly different in efficacy from the most effective treatments (viz. those completely inhibiting leaf-spot development) was re-tested (Table 2). To these were added materials under certification based on the same active ingredients as the effective formulations in Trial 1. In the final selection, tests were made with the previously most effective materials (Table 2).

Finally, rain resistance of successful treatments was tested approximately 24 hours after application by spraying the plants for 6 minutes on the turntable with water equivalent to one inch rainfall (4.5 gal. per sq. yd.). Results of this test are given in Table 3.

TABLE 3. TRIAL 3. EFFECT OF SIMULATED RAIN ON MATERIALS FOUND SATISFACTORY IN THE FINAL SELECTION

Treatments	Amount (lb.) in 100 gal. water	Lesions per leaf stalk	
		Mean No.	Log (x + 1)
Check	—	295.2	2.45
Fermspray	4	0.0	0.00
Fertram	3.5	1.0	0.22
Tetram	4	8.3	0.89
Thiotox	4	7.0	0.72
Thirospray	3.2	11.6	1.03
Dithane D14	2 : 1 : 0.5	0.0	0.00
Manzate	2	21.6	1.19
Phygon XL	0.25	2.8	0.43
Flit 406	2	9.0	0.94
Orthocide	2	21.2	0.78
Nirit	5	139.8	2.09
Karathane	2	114.4	1.94
Difference required for significance at 1%			0.57

RESULTS

Under these test conditions, materials can be grouped according to the measure of control obtained.

Leaf-spot infection under conditions of artificial rain (Table 3) was effectively prevented by: ferbam at 0.28% (Fermspray at 0.4% and Fertram at 0.35%); zineb in a tank mix formulation of nabam at 0.038% plus zinc sulphate at 0.1% (Dithane D 14 at 0.2% + zinc sulphate at 0.1% + hydrated lime at 0.05%); and dichlone at 0.013% (Phygon XL at 0.025%).

Satisfactory control was obtained with the following materials in Trials 1 and 2 (Tables 1 and 2): ferbam at 0.28% (Fermspray at 0.4% and Fertram at 0.35%); zineb at 0.138% (tank formulation of Dithane D14, nabam at 0.038% + zinc sulphate at 0.1%); maneb at 0.14% (Manzate at 0.2%); dichlone at 0.013% (Phygon XL at 0.025%); captan at 0.1% (Flit 406 and Orthocide at 0.2%); dinitro benzene at 0.075% (Nirit at 0.5%); and dinitro phenyl crotonate at 0.05% (Karathane at 0.2%).

In general, thiram prevented leaf-spot infection, but formulations varied in efficacy, thus Tetram at 0.4% (0.32% thiram) was effective in 2 out of 3 tests; Thiotox at 0.4% (0.32% thiram) was effective in 1 of 2 tests and Thirospray at 0.32% (0.16% thiram) was effective in 2 tests at half the concentration of either Tetram or Thiotox.

Results with ziram at 0.14% (Zirospray and Fuclasin Ultra at 0.2%), and lime sulphur at 0.75% were inconsistent.

Glyodin at 0.136% (Crag 341 C at 0.4%) gave satisfactory control in 2 out of 3 tests but caused plant damage.

Chloranil at 0.012% (Spergon w.p. at 0.025%), and zineb at 0.13% used in a proprietary formulation (Dithane Z78 at 0.2%) were effective in 2 out of 3 tests and 1 out of 2 tests respectively.

The following materials, at concentrations tested, were not as effective as the above: copper compounds, sulphur with exception of lime sulphur at 0.75%, salicylanilide, and nitro benzene dusts.

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